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A modest step forward in New York

IN 1969 the United Nations solemnly declared the next ten years a Disarmament Decade. Now, as this period draws to a close, one may reasonably wonder whether it has been anything other than meaningless, with so little progress on so many fronts. Perhaps the only chance that these ten years will be remembered is that the United Nations General Assembly's special session on disarmament, recently ended, will prove to be a turning point. And on that, one can at least say that it didn't end up as a disaster; that a final document of substantial proportions was agreed on; that measures have been taken to keep disarmament nearer the centre of the international stage; and that although nothing spectacular has emerged, there was an air of quiet satisfaction about many of the participants.

It is forty-seven years since the last truly international conference on disarmament, although of course there have been all manner of bilateral and multilateral gatherings. Thus there was a certain amount of trepidation about this giant assembly. Would it degenerate into pious talk, abuse or just airy nothings? In the end these worst fears were not realised. Certainly the final document is overinflated and there are the compulsory remarks about the new international economic order, general and complete disarmament, the armaments of racist regimes and so on. But on the whole there is ample evidence that most participants understood the excruciating complexity of balancing security with disarmament.

If there is one major criticism to be directed at the report it is the obsession with nuclear weapons. They are declared top priority in disarmament negotiations and they are called the greatest danger to mankind and to the survival of civilisation. This perfectly proper unhappiness with the dilatory way in which nuclear-weapon states have approached disarmament has distracted just a little too much attention away from conventional arms and the massive trade in them. The strongest remarks the assembly can muster on this latter subject are that "consultations should be carried out among major arms supplier and recipient countries on the limitations of all types of international transfer of conventional weapons . . . with a view to promoting or enhancing stability at a lower military level, taking into account the need of all states to protect their security as well as the inalienable right to self-determination and independence of peoples under colonial or foreign

domination . . .". Such tepidly recommended consultations seem most unlikely ever to come about. Many would have liked to see the UN generate a really major study on the arms trade, but, it is said, India led a group of nations which blocked any such development.

Most of the proposed projects and studies got politely shunted off into an appendix. One that did, however, receive favourable mention was on the relationship between disarmament and development. The Secretary-General of the UN has been charged with convening a study group to prepare a 'forward-looking and policy-oriented' report on this issue.

It was widely expected that the present Conference of the Committee on Disarmament which meets in Geneva would not survive intact, and so it has turned out. The present arrangement of 31 countries, under the co-chairmanship of the United States and the Soviet Union, has not been entirely satisfactory, particularly because China is excluded and France has chosen not to show up. In the new arrangement the Committee on Disarmament will be up to 40 in number, and the chairmanship will rotate throughout all the members; there is a clear expectation that this will bring France, and eventually China into the fold. The assembly also made a distinction between a gathering to negotiate (which is what the Committee on Disarmament is) and a gathering to deliberate. For the latter purpose a Disarmament Commission of all members of the UN has been set up. This commission will, amongst other things, follow up the decisions and recommendations of the special assembly.

If nothing remarkable has emerged from this special session, perhaps the most tangible advance has been the 'negative security assurances' that nuclear-weapon states have made public. These are undertakings not to use nuclear weapons against non-nuclear states which are not acting in conjunction with a nuclear state. The United States, Soviet Union and United Kingdom have given such assurances; China has given a no-first-use pledge and France has simply declared she will respect nuclear-weapon-free zones.

Delegates left New York promising that the special session would be repeated, perhaps in three years time. By then SALT 2, a test ban and maybe further nuclear-free zones should have been negotiated. The next step must then be to face up to trade in conventional weapons. □



David Dickson reports on the growing strength of the anti-nuclear movement—and a shift in strategy at the Nuclear Regulatory Commission.

THE US anti-nuclear movement received a major morale booster last week when the Nuclear Regulatory Commission ordered the indefinite suspension of construction work on a 2,300 MW nuclear power facility at Seabrook in New Hampshire.

In what opponents of the facility hailed as an "historic breakthrough", and the state governor labelled an "asinine decision", the NRC commissioners voted by two to one to suspend construction until adequate studies of alternative sites have been carried out.

The decision was essentially procedural rather than technical. It followed a ruling by an appeal court in Boston in February that the Environmental Protection Agency had followed incorrect procedures in giving its approval to a cooling system which would discharge heated water into the sea (thus, according to critics, posing a threat to local marine life). In addition the NRC's Atomic Licensing and Safety Appeal Board ruled in April that not enough thought had been given to alternative sites for the power station, given that the choice of cooling systems was still open.

The EPA is holding new hearings into the proposed cooling scheme. The decision that faced the NRC commissioners was whether, in the light of these hearings and the appeal board ruling, construction should continue while the matters remain unresolved. Two of the three commissioners—physicist Victor Gilinsky and lawyer Peter Bradford—decided that, in view of the uncertainties, the suspension of work was justified.

The decision will mean that almost 2,000 workers will be laid off by the Public Service Co. of New Hampshire which began construction of the \$2.3 billion facility in 1976, and says that it is now 10% complete.

The two commissioners stated that the lay-offs were "the factor which weighed most strongly against suspension". However, they added: "We can only say that the opposite course would cause greater harm through failure to comply with the law and would risk the same impact on the workers through a court-imposed injunction in the immediate future."

The NRC's decision was announced at the end of a week which had seen a four-day rally attended by many thousands of protesters at the construction site in New Hampshire, and a subsequent three-day demon-



Anti-nuclear protesters at Seabrook

US regulators tread more warily on nuclear power

stration outside the NRC's offices in Washington by 100 members of a group called the Seabrook Natural Guard.

The decision itself came as a surprise to many of the demonstrators, who distrusted the NRC on the basis of previous pro-industry decisions. It was only after considerable debate within the Clamshell Alliance, a New-England-based collection of 53 groups which has been coordinating the Seabrook protest, that the decision to stage a Washington demonstration was made at all.

One effect has been a psychological boost for the anti-nuclear movement, which has been slowly gathering strength over the past few years, and whose conscious commitment to civil disobedience is reminiscent of the UK's Campaign for Nuclear Disarmament in the early 1960s. Following demonstrations at Seabrook over a number of years, with 180 people arrested in August 1976 and 1,415 during an occupation of the site in May 1977, similar organisations have sprung up across the country in opposition to local nuclear installations.

Seabrook, however, has remained the centre of attention. During the New Hampshire demonstrations, many of the other groups held supporting rallies. And although so far there has been a reluctance to bring grass-roots movements to Washington, a number of environmentalists feel that major demonstrations in the nation's capital may now become more common.

It is difficult to tell whether the demonstrations had much direct influence on the NRC's decision to suspend construction at Seabrook. But pressure from environmentalist groups has certainly played a major part in bringing decision-making into the open.

The NRC has become increasingly sensitive to criticism from both the public and Congress so that, despite its

institutional separation from the Atomic Energy Authority in 1975, it has continued to reflect a desire to protect the nuclear industry.

Mr Russell Peterson, director of Congress' Office of Technology Assessment, expressed such concerns when he said recently that postwar strategies to develop civilian nuclear power had been "seriously flawed", and that inadequate efforts to insure the safety of nuclear power meant that "rather than enjoying wide public confidence, it has become highly controversial".

In recent months, both the commissioners and NRC staff members have been attempting to change their traditional image, recognising that, as commissioner Bradford commented on the Seabrook decision, "the courts, the legislative bodies, and the public are unlikely to tolerate nuclear expansion unless the regulators take the laws and their duties seriously".

Many have interpreted the Seabrook decision as a step in the right direction. In the past, for example, the NRC went to great lengths to keep a facility from shutting down. "Now at least it seems as if the commission is leaning over backwards to be procedurally fair," says Professor Frank von Hippel of the Centre for Environmental Studies at Princeton University.



Commissioners Bradford and Gilinsky

Further evidence that the NRC is pursuing a more independent role than in the past is reflected by the increasing criticism—previously reserved for agencies such as the Occupational Safety and Health Administration—that it is beginning to attract. Following the Seabrook decision, for example, Congressional representatives from New Hampshire called for legal action to reverse the ruling. They want to amend the Department of Energy and the NRC appropriations bills to assure construction of the plant.

It would be wrong to interpret last week's decision as indicating any major weakening in the administration's support of nuclear power. A number of other recent decisions—such as the Supreme Court's ruling that the Price-Anderson Act, which limits public liability for nuclear accidents to \$500 million, is not unconstitutional—have not helped the anti-nuclear cause.

Furthermore even minor victories can have their price. Last week, a number of staff members responsible for a Congressional report critical of the way that subsidies had distorted the apparent costs of nuclear power were fired from their posts.

Yet whether or not the NRC allows the Seabrook plant to go ahead, both sides now recognise that nuclear power has become a political issue, with many wider issues at stake than merely the well-being of a few shell-fish—or the adequacy with which government regulators fulfil their functions.

During the Seabrook demonstration, for example, Dr John Goffman, for many years a critic of the health dangers of nuclear energy, told the rally that "nuclear power is a symptom of a societal disease, the existence of privilege and power", a theme echoed in many other speeches.

Equally Governor Meldrim Thomson of New Hampshire has vowed to make Seabrook the rallying cry of a pro-nuclear, grass-roots movement; while a research fellow from Harvard University, criticising the protesters in a letter to the *New York Times*, complained that "to protest nuclear power is to strike at the marriage of science and capitalism".

As far as Seabrook itself is concerned, the ball now rests with EPA administrator Doug Costle, to whom environmentalists are pressing their case as he decides whether to give the planned cooling system the go-ahead.

As to the broader issues, with regulatory action now making a major economic impact on the nuclear power industry and the NRC taking an increasingly independent line, the struggle between the supporters and the opponents of nuclear power seems destined to grow more intense. □

Lack of basic energy research criticised

THE long-range security of US energy supplies has been jeopardised by the failure of the Department of Energy to give sufficient attention to long-range, fundamental research, according to a study commissioned by the Office of Science and Technology Policy.

The working party which carried out the study recommends that the balance between short-term and long-term needs in the department's research programmes "should receive early attention at the highest levels".

The working party also says there is a lack of balance in the research programmes carried out on federal funds by DoE laboratories, industry and the universities. To redress the balance, it suggests the budget for basic energy research in the universities should be increased. If necessary "funds should be diverted for this purpose from the development activities or even from other parts of the basic research programme".

The study was commissioned by Dr Frank Press, director of OSTP, in December 1977, and was carried out by a working party that included representatives of industry, government and the academic community, chaired by Dr S. J. Buchsbaum of the Bell Telephone Laboratories.

In a scaled-down version of comments which OSTP has itself made of federal research efforts in general, the working party criticises a preoccupation with near-term programmes and the consequent neglect of longer-term work. The dearth of research is especially evident, it says, in the solar and fossil fuel programmes.

Among the reasons for this imbalance, the working party lists a "misguided emphasis" on goals given to, or adopted by, the department which are clearly not yet attainable, and "excessive sensitivity" of some programme managers to political pressures.

As a mechanism to correct this situation, it is suggested that a committee be established under the

chairmanship of the director of the Office of Energy Research, which should decide how to allocate funds to basic research.

Under this structure basic research would remain the responsibility of each assistant secretary, and a linkage therefore maintained between the research and its applications; however it would be less easy for a particular division to claim that it had increased its basic research activities merely by redefining existing programmes.

As far as federal laboratories with an applied research mission are concerned, the working party does not make any specific recommendation. But, picking its words carefully, it says that the "crispness of their missions has slowly eroded" in recent years, and that there exists a need for "greater quality control" over their basic research effort.

Turning to specific research needs and opportunities, the report lists a number of areas—including fossil fuels, fusion energy, and large scale solar power—in which it says long-range fundamental research should more frequently be given priority. As far as research in the environmental and life sciences related to energy matters is concerned, the report says it is essential for the DoE to avoid the "credibility problems" of its predecessor agencies. It therefore suggests that primary responsibility for basic research should be assumed by the director of the Office of Energy Research where the work is common to several energy technologies, and that the Assistant Secretary for the Environment should provide support for basic research relevant to environmental concerns in the technology area.

Although the report has no official status, it is said to have been well received both by Dr Press, and Energy Secretary Dr James Schlesinger. Its reception by both fiscal conservatives, and those Congressmen concerned with specific development projects, however, is less predictable.

David Dickson

Decline in scientific knowledge among 17-year-olds

THE science scores of 17-year-olds in US schools has dropped by 4.7% since 1969, according to a report released in Washington by the National Assessment of Education Progress.

A study of 9-year-olds and 13-year-olds showed less of a decline and an improvement in some areas. For example, both 9 and 13-year-old students improved in tests in the biological sciences.

However in all three age groups scores in the physical sciences were

lower in the 1976-77 school year than in either of the previous two examination periods in 1969-70 and 1972-73.

The declining performances of 17-year-olds seems closely related to the fact that the number of students taking science courses in high school has dropped—from about 18% in the late 1960s, when attention on space exploration heightened interest, to less than 10 per cent now. □

Can ESA members afford to fly on Spacelab?

THE UK and several other member states of the European Space Agency are having doubts about the cost of flying experiments aboard future missions of Spacelab, the European-built space station which will be carried into orbit in the cargo bay of the US Space Shuttle. Response to ESA's call for experiments in April has been so good that if all the UK proposals (about 25) were recommended for flight (an admittedly unlikely eventuality), the total cost would come to perhaps double the £25 million annual outlay of the Science Research Council for all its astronomy, space, and radio activities.

In practice, preliminary ESA payload breakdowns are allowing the UK 13–14% of the Spacelab payload, but even this more realistic slice of the action would cost approximately £3 to £4 million per flight. Unless more money can be released from the research councils or the Department of Industry, the UK may be unable to afford this much. France, which is being offered a slightly greater percentage of the payload, is also expressing reservations about flight costs, as are Italy and the Netherlands. But Germany, which is largely responsible for the construction of Spacelab, seems to have no such worries, and has agreed to provide half the payload of the four missions being discussed.

These four European missions will follow the initial flights of Spacelab which have already been agreed. Payload space for Spacelab's first flight, now unofficially expected in April 1981, has been split between NASA and ESA, and NASA is offering a free launch. Spacelab 2, an American scientific mission, has some UK experiments on board, but these are being flown free as a cooperative venture. For the follow-on Spacelab missions, currently under discussion, the European experimenters will have to pay for the launch as well as the payload, and it is at this stage that several ESA member states are beginning to wonder what they have let themselves in for.

Two of the missions, referred to as D1 and D4, are being sponsored by the Germans, who are inviting other experimenters to join them on board. The other two missions, termed DM (Demonstration Mission) 1 and 2, are being paid for by ESA and the costs

are being shared among the individual experimenters. Launch schedules will be clarified during the next three months, but as currently envisaged the German D1 mission will probably be the first to fly in June 1982. This will be a microgravity mission, concentrating on experiments on materials science and life science in weightlessness. Then will follow DM2, an Earth-oriented mission, in autumn 1982, with another micro-gravity mission, DM1, in mid-1983. Last, because of its complexity, will be the German astronomy mission D4, probably in late 1983.

The SRC is now deciding which UK experiments it can support; part of its job has been to find sponsors for experiments in areas such as materials science and life sciences that do not come under its jurisdiction. The number of proposals vying for consideration is likely to drop as experimenters with common interests combine their activities to share costs, but there are bound to be some proposals that would be too big, too heavy, and thus too expensive, to gain support. Since most of the SRC's limited budget is already earmarked, the Council is unlikely to be able to afford more than a few million pounds for Spacelab experiments, meaning that it could support a total payload weight of only around 200 to 300 kg for the four missions.

One possibility is that the Department of Industry, which has so far paid the UK's 6.3% contribution to the cost of building Spacelab, might be persuaded to continue its support for Spacelab past the development stage, thus paying the boarding costs of experiments and leaving the research councils with only the funding of the actual experiments to worry about. But the drawback to this from the DoI's point of view is that relatively few experiments have so far been proposed by British industry, most of the response having come from universities and research establishments.

Ian Ridpath

Space medicine: a Polish speciality

DURING the 12 years of the *Interkosmos* programme of space cooperation between the countries of the socialist bloc, space medicine has become a Polish speciality. Not surprisingly, therefore, the flight of Mirosław Hermaszewski as part of the current "international" manned-flight programme of *Interkosmos* included a number of medical experiments.

These ranged from "zdrowie"—an estimation of physical efficiency by means of simultaneous ECG analysis, monitoring of the rate of cardiac con-



Polish cosmonaut Hermaszewski

tractions, blood pressure, respiration rate and body temperature—to the somewhat exotic "smak" experiment, to elucidate the "still unclear mechanisms of the taste disorders experienced by cosmonauts in orbit". The latter, which was contributed by the Polish Military Institute of Aviation Medicine, is based on the electrical stimulation of the taste buds.

The programme also included the "relaks" experiment, which estimates the influence of "recreation exercises" on the psychophysiological condition of the crew in the course of a prolonged flight, and the "kardiolider", which monitors cardiac strain to determine the physical effort exerted by a cosmonaut in various stages of the flight. The latter device, developed by the Warsaw Institute of Aviation Medicine, is said to be of particular use in monitoring physical exercises for cosmonauts in conditions of weightlessness, as well as their readaptation to the earth's gravitation on their return.

Stressing the contribution of the non-Soviet crew member to an "international" flight is, of course, important for purely diplomatic reasons. In the first such flight, earlier this year, considerable emphasis was placed on the Czechoslovak contribution—notably the Czech capsule "morava" which was used in conjunction with the Soviet "Splav" furnace as part of a series of experiments producing alloys which would be impossible under conditions of gravity. (The present mission included a similar joint experiment, with a Polish capsule code-named "syrena").

What is remarkable about the medical experiments is that both sides stress that they are predominantly a Polish contribution. According to veteran cosmonaut Vladimir Aksenov, interviewed on Moscow radio, the Poles are developing the experiments begun in the Soviet Union using their own methods and their own instruments.

Vera Rich

Shcharanskii trial stirs new anti-Soviet protests

SCIENTIFIC and technological cooperation should not be linked to questions of human rights and the implementation of the Helsinki accords, according to a Tass statement issued last month in the aftermath of the Orlov trial. This sentiment is clearly not reciprocated abroad. The trial of Anatolii Shcharanskii, cyberneticist and human rights campaigner, this week has already evoked a wave of protest.

More than 200 French scientists are petitioning for a boycott of scientific contacts with the Soviet Union, in protest against the sentence on Yurii Orlov, while two high level American visits by Dr Frank Press, President Carter's scientific adviser, and Barbara Blum, deputy administrator of the Environmental Protection Agency to the Soviet Union have been cancelled because of the Shcharanskii trial.

The Soviet Union is clearly extremely sensitive to the cancellation of such scientific contacts. One of the

charges made against Shcharanskii in the Soviet press at the time of his arrest last year was that he had been working towards the disruption of scientific détente. Describing the alleged espionage activities of Shcharanskii and his associates in the human rights movement, *Izvestiya* stated: "A letter from Rubin arrived August 1976 through unofficial channels via the American correspondent Osnos. It requested a quicker forwarding of this information so as to start a campaign to put a ban on the sale of American equipment to the USSR."

According to his accusers, therefore, Shcharanskii was not simply guilty of espionage: his offence was exacerbated by being directed against the development of scientific and technical exchange. It would be ironic, indeed, if his trial resulted in a complete or partial breakdown of such contacts.

However, a complete boycott of all scientific exchange seems unlikely—if

only for the pragmatic reason that, if the ultimate weapon is unsuccessfully deployed, there are no further bargaining resources.

Dr Robert W. Kates, chairman of the Committee for Human Rights of the US National Academy of Sciences, stresses the "remarkable response on the part of the American scientific community" in defence of human rights activists such as Orlov and Shcharanskii. Already, he said, there had been many voluntary and individual acts of conscience. He further noted that while scientific exchanges can be agreed at official levels, participation in them is voluntary. "Science is a voluntary activity".

He feared that, as a result of the Shcharanskii trial, "there will be a deep and persistent sense of disillusion" among scientists. This would continue unless there was "some sign of a judicious and humanitarian response by the Soviet authorities". □

It's all in the mind . . .

ACADEMICIAN Andrei Snezhnevskii, Director of the Institute of Psychiatry of the Academy of Medical Sciences of the USSR has become well-known of recent years in the western press—not the least for his remarkable discovery of "sluggish" or "creeping" schizophrenia—a mental disorder which presents no clinical symptoms.

Not surprisingly, western specialists have become intrigued by this illness, which seems confined entirely to the Soviet Union—where however, it seems endemic among a certain section of society.

For, in particular "sluggish" schizophrenia (a subdivision of "continuous") is a particularly powerful concept in Soviet forensic psychiatry. It is, in the words of one prominent Soviet psychiatrist, "schizophrenia without schizophrenia". It is expressed only in certain behaviour patterns, showing "poor adaptation to the social environment". In a handful of cases, this has been invoked for such deviations as a hippy-like mode of dress. The typical "sluggish schizophrenic", however, exhibits "paranoid delusions of reforming society"—or dissidence and a concern with human rights.

Last week, Dr Ilya Glezer, a former member of the staff of Snezhnevskii's institute, who is now working at the University of Beersheva (Israel) visited London, providing an opportunity for *Nature* to learn more of Snezhnevskii's principles of diagnosis.

Dr Glezer is himself not a psychiatrist but a neurocytologist. In general,

Soviet psychiatry stresses the physical and genetic background of disorders and de-emphasises the environmental factors. As part of his post-doctoral research, Dr Glezer was to prepare a survey of post-mortem brain analyses of deceased psychiatric patients.

On commencing his research he was instructed to pay great attention to the case-histories of the patients. Accordingly, he delivered the case-notes to a team of close associates and pupils of Academician Snezhnevskii, who pencilled on each the diagnosis according to the director's scheme "continuous schizophrenia", "periodic schizophrenia", "shift-like schizophrenia". (It should be noted that to Snezhnevskii, all mental disorder can be classified as a form of schizophrenia.)

After completing his investigation of some fifty cases Dr Glezer was ready to present his findings. Again he was told to check up on the case-histories. Some time having elapsed since the first classification was made, he carefully rubbed out the first set of diagnoses, and returned the notes to Snezhnevskii's team. Later the notes came back—with different diagnoses.

Since the patients concerned were all dead, Dr Glezer found himself unable to explain this phenomenon—except on the assumption that the various types of schizophrenia of Snezhnevskii's system do not correspond to any defined clinical pattern, but are assigned more or less arbitrarily. (Since the "diagnoses" were made by



"And remember, comrades, illness after death does not prove life after death"

some of Snezhnevskii's closest associates, one can hardly impute to them incompetence in applying Snezhnevskii's criteria).

In due course, Dr Glezer was dismissed from the institute and later sentenced to three years in prison and three years exile, on the charge of slandering the State. The formal evidence against him was the writing of letters to government and Party officials in support of the Jewish emigration movement.

Dr Glezer readily admits writing such letters. However, many other Jewish activists and would-be emigrants have written similar letters—without incurring such harsh sentences. The suspicion inevitably arises that he was jailed not only for the letters but also of his exposure of the arbitrary nature of Snezhnevskii-type diagnosis.

Vera Rich

HSE report offers little advice to UK universities

THE report which everyone concerned with health and safety in UK universities has been waiting for for almost a year was at last published by the UK Health and Safety Executive last week. Its publication, however, could come as something of an anti-climax. Some academics had expected that it would give the universities a clean bill of health and therefore embarrass the unions; some unions, had expected that it would find fault in university practice and so embarrass some academics. But the report seems to pass little judgement on the state of health and safety in the universities especially where research is concerned. It is primarily a description of the hazards found in universities and the ways in which the universities go about controlling them.

'Working conditions in universities', is the outcome of a pilot study of six selected universities carried out in 1976-77 by Miss N. Curry, a senior inspector of the Factory Inspectorate, and by inspectors of the Nuclear Installations Inspectorate. The study was initiated by the HSE to provide an assessment of the hazards found in universities as a guide to its inspectors. This first report, however, has been prepared mainly to provide background for further discussions between employers, unions and the HSE before the HSE decides what action to take.

Although the report is largely descriptive, it does find subjects for criticism and praise. Greatest fault is found not by Miss Curry but by the nuclear installations inspectors. Tucked

away in appendix 9 at the back of the main report is the report of their survey on radiological protection.

Standards are low, they say, and in many cases fall below the minimum standards laid down by guidance notes which have been in force for many years. They cite examples of barriers and warning notices being totally ignored; of adhesive tape labelled 'radioactive' being used indiscriminately; of inadequate monitoring of the working environment for radioactivity and of food being stored with radioactive materials. They also report that precautions taken when working with x-ray sources and lasers were often inadequate.

Some of this poor state of affairs they attribute to a lack of coordination because the responsibilities of radiation protection officers are not properly defined. As part of the solution they suggest that each university should have its own central organisation which would make routine visits to different departments to check that individuals are taking the proper precautions.

For Miss Curry, however, the greatest danger to university workers seemed to stem from the poor design of buildings about ten years old. Even though she says that research work is "possibly potentially the most hazardous function of a university" she feels that the risk involved in maintaining building and plant is greater than that associated with research.

A list in the report of some typical hazards associated with research in-

cludes compressed and liquified gases, carcinogens, acids, dust, microbiological hazards, electrical hazards and noise. In each case a brief description of the hazard is given but little indication of whether the HSE feels it is being controlled adequately or not. Possible areas for improvement, however, are the disposal of hazardous wastes and the ventilation of preparation rooms.

In general, Miss Curry feels that all ancillary processes in the universities should be conducted in accordance with regulations laid down for factories. In teaching and research laboratories, however, she says, this may not be possible. So far as the use of highly flammable liquids and gases is concerned, regulations for factories may not be appropriate and the report suggests that a code of practice for the storage and use of highly flammable material should receive early attention.

The next stage is for the HSE to collect opinion on the report and then presumably to formulate guidelines or recommendations. In the meantime the Factory Inspectorate is to start an inspection of all universities in the UK. At the rate of 20% a year, it plans to have inspected them all by 1982. The report, however, is unlikely to give safety advisers and officers much clue as to what the HSE inspectors will now be looking for.

Judy Redfearn

Working conditions in universities. Report on a survey by Miss N. Curry available from the Health and Safety Executive, Baynards House, 1 Chepstow Place, London W2 4TF, price £1.75.

Company's claim sparks fresh controversy over Seveso

DISCUSSION of the accident which occurred in the trichlorophenol reactor at Séveso in 1976 still tends to result in heated debate. This was illustrated during a recent New York Academy of Sciences conference on halogenated hydrocarbons.

Dr Francesco Pocchiari of the Istituto Superiore di Sanita in Rome was scheduled to speak on the health of the population exposed to 2,3,7,8-tetrachlorodibenzo-p-dioxin (dioxin) at Séveso. On the same day, the *New York Times* printed an exclusive story about an internal newsletter of Hoffmann-La Roche (parent company of Givaudan, owners of the factory at Séveso) with the headline "Company says '76 blast in Italy caused little injury".

Several of the Italian representatives at the meeting felt the information was perhaps intended to cut the ground from under Pocchiari's feet, before he

could present his report—virtually on behalf of the Italian government. Pocchiari reported that 187 cases of the skin disease chloracne had been confirmed following the screening of 32,200 children in the Séveso region; that the severity of the skin lesions was lessening; that some peripheral nerve damage had been detected in adults in the area; but that there was no increase in foetal abnormality which could be linked with certainty to dioxin exposure. Several of the investigations had their limitations and Pocchiari indicated that his paper must be seen as a progress report.

At a press conference, Pocchiari said Roche's action in making their claim was "premature". "If the dioxin was carcinogenic, as several studies now suggested it was, is it not the long term health effects we ought to be considering?" he asked. Dr Cesare Maltoni of the University of Bologna,

a recognised authority on the carcinogenic action of vinyl chloride, echoed these views.

The Roche newsletter does, however, mention the possibility of "belated consequences" to the health of the residents, which "cannot be ruled out". But it adds that scientific findings allow "the confident assumption" to be made that Séveso residents were exposed to small amounts of dioxin, and that no "serious and permanent" damage to health was sustained.

Roche's representative at the conference was caught unawares by the *New York Times* story. He was told about it by a questioner. As for the *New York Times*, they claim their story was filed on the June 20 and printed on June 25 simply because of previous lack of space.

Alastair Hay

Pakistan calls for new international institutions

PAKISTAN'S draft country paper for the UN Conference on Science and Technology for Development has now been circulated to various science bodies in Pakistan by the Ministry of Science and Technology. The 35-page document is a result of a long debate among the various scientific associations and government agencies in Pakistan, so the final version of the paper will probably not differ significantly from the views expressed in the draft, which lays great stress on international co-operation.

The UNCSTD conference has three main points on its agenda:

- Science and technology for development;
- Institutional arrangements and new forms of international cooperation in the application of science and technology;
- Utilisation of the existing UN system and other international organisations.

The draft has reviewed these issues as they affect Pakistan and the rest of the world, and has made some concrete proposals for improvement.

Pakistan, like most developing countries, faces great internal social disparities. So far the development pattern has been such that the elite and the urban areas have gained more than their due share, while the rural and the depressed sections have been relatively deprived from such benefits. The result has been great social unrest. But the country is striving to create a new socio-economic order wherein the benefits of development should be available to all.

The draft country paper distinguishes five priorities: water management; industrialisation; scientific and technological manpower; natural resources; and energy. These are rated as the most crucial for Pakistan. The draft also discusses technology transfer, the better utilisation of technical knowledge, and new science and technology.

Pakistan's experience of technology transfer has been far from satisfactory. Such transfer has mostly been through licensing or through joint ventures; direct investments have been rare. Technology transfers have largely been package deals on a turnkey basis. Such horizontal transfer is capital intensive (in foreign exchange), labour saving, and ultimately does not lead to grass roots development. Further, restrictive clauses in technology-transfer contracts — such as restrictions on exports, on the pattern of production, and so on, have held back proper and rapid development. In the distribution of licence agreements, the emphasis has been on consumer goods, to the

detriment of capital equipment. The draft puts forward a number of suggestions to correct this imbalance.

So the draft makes a proposal that aid from UN and other international organisations should be made available to set up institutions for technology assessments which could assist local entrepreneurs in making a proper choice of technologies and in negotiating more reasonable terms in licensing agreements.

Some other significant points in the draft are:

Institutional arrangements for irrigation and water management are weak. Existing irrigation research institutes need strengthening in manpower as well as equipment through the help of international bodies. There should be international assistance in using sophisticated techniques like remote sensing from space for better water manage-

priorities is needed at the global, regional and project level by the UNDP Technical Assistance Programme. A fund should be set up by UNDP for financing and strengthening the Technical Cooperation among Developing Countries (TCDC)".

The developed countries should "also be urged to contribute their due share of at least 0.3% of their GNP towards UNDP. In addition, developing countries with *per capita* income of more than \$400 may also be asked to contribute to this fund".

The draft continues: "in devising the mechanisms for future international cooperation, the paramount consideration should be the principles on which the Declaration of the Establishment of a New International Economic Order is based. Following from these, the major thrust for international cooperation and concerted

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Proper water management is a priority

ment. Technically trained and reliable manpower in fields like electronics, metallurgy, industrial design, petrochemicals, nuclear technology is lacking. To rectify this there should be increased international support to universities in such disciplines.

Exploitation of natural resources, particularly in mining and agricultural sectors is very weak. Institutions in these sectors need international support for R&D.

Regional collaboration ought to be encouraged among developing countries in non-conventional energy technology, since it is likely to be a common factor in the economic development of the whole equatorial and sub-equatorial belt.

Scientific and technological research calls for a heavy investment of capital and human resources, and no developing country can mobilise them on the required level. International cooperation is essential. Individual countries have entered into bilateral agreements with friendly countries or have formed regional cooperative bodies but a "major shift in the emphasis and

action will be to assist the developing countries in achieving a large measure of technological self-reliance through the building up of adequate indigenous science and technology capabilities for solving their problems of development and for working out a system through which they can benefit from each others capabilities and expertise, reducing dependence on developed countries."

As a corollary, it has been suggested that a network of cooperative institutions must be set up in the developing countries to promote a joint research effort on common problems. Some of these institutions with exceptional potential should be graded as "centres of excellence" to be supported by the UN.

The communication of scientific information also needs to be overhauled, says the draft. There should be a single focal point at the UN level instead of the multiplicity of different UN agencies each providing their own information. Private institutions should also be encouraged to provide information on their technologies.

Azim Kidwai

correspondence

Greek universities

SIR,—I would like to respond to some of the remarks of Dr Pantelouris on Greek universities (2 February, page 395).

We select students by written examination and everyone with 12 years schooling is eligible. The applicants (80,000) are almost all of those who have finished high school, regardless of their marks, as every father wants to see his child in university. About 15,000 get to university and another 15,000 to vocational colleges. Dr Pantelouris excludes educational colleges for elementary school teachers. About 40% of high school graduates get into higher education. This number compares very well with other countries. 85% of the successful applicants pass their entrance exams first time. Tutoring is necessary for students with low marks, but not for the better students. Unfortunately many worry and unnecessarily attend tutoring classes.

Some parents want to see their children enter a university so much that they send their children to Italy, although many return without a degree. This is a social problem, as we do have a great number of students who study abroad, receive a degree and then return expecting a life time job. It is hoped that the reforms in secondary education (presently underway) will meet this problem.

"Lectures are formal and *ex-cathedra*" says Dr Pantelouris. As far as I know lectures are formal in all universities. I think your correspondent does not read books on education. Research done in the US informs us that "student and faculty performance—whether in interdisciplinary and team courses, student-centred curriculum, written-evaluation grading or any other structure—proved to be much the same in each programme examined" (A. Levin and J. Weingart, *Reform of Undergraduate Education*, Jossey-Bass, 1973). Therefore the criticism of Dr Pantelouris is not valid. However, our students do participate in class discussions and seminars. We follow the progress of the means and methods of the teaching and we try to apply them.

Since 1975 every one of our staff who holds a Ph.D. has the right to teach, and it is up to the faculty to decide who will teach what. Here, Dr Pantelouris misrepresents the case. The law obliges the full professor to teach the main course and leaves other courses to be taught by the young professors. So the law protects the quality of teaching and also avoids mistakes already made in other countries. Dr Pantelouris writes that there is one full professor per unit. As far as I know, the same holds for British universities. He then goes on to speak about "the rest of the staff" who have just a first degree. It is a mistake to consider them "staff", as they are students working for their Ph.D. degree and are selected as demonstrators. For this purpose they take an assistantship lasting six years, possibly nine. Some universities consider them as instructors. He then continues "there is nothing comparable to the British 10:1 student-staff ratio. No average figure can

be given, but however calculated, the ratio would be several times higher". I have just calculated that for our school it is 5:1.

Dr Pantelouris thinks that our internal Ph.D. degree is granted on the discretion of the professor and not on the research ability of the candidates. This is quite wrong as it is granted by the faculty of the particular school, where the candidate has to defend the thesis.

His remarks on bureaucracy are correct. However this is a problem for universities all over Europe; some are better and some worse than we are. Dr Pantelouris offers advice on what we have to reform. Theoretical knowledge is not enough for giving advice. Living and experiencing are important factors for offering constructive criticism and to avoid destruction of that which stands correct. What is good for the Greek universities, is not easily determined. There must be careful investigation and research work before anybody tears down the existing system.

Dr Pantelouris should know that attempts at reform, however stimulating and numerous and creative and hopeful, are at loggerheads almost everywhere with traditions—traditional student passivity and traditional university reward systems that extol specialisation and concentration.

G. A. MOURKIDES

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Thessaloniki, Greece*

Conference ethics

SIR,—In our letter (16 February, page 605), we described our unsuccessful attempt to attend the Fourth International Meeting on Ferroelectricity held in Leningrad 18–23 September 1977. The response of I. S. Zheludev (20 April, page 666) is of interest and deserves a reply.

First, Zheludev claims that had Havlin merely applied to the Soviet Embassy in Rome, he would have found his visa waiting for him. The only formal response by the Soviet organising committee to all of our letters was sent to the International Union of Pure and Applied Physics (IUPAP). In early July 1977, the Secretary of the Israel Physical Society, acting on Havlin's behalf, received the following cable from Larkin Kerwin, Secretary General of IUPAP: "I have just received from Leningrad following telegram: 'Professor Havlin's paper is included in the meeting programme. He should apply for a visa at Intourist or to a firm acting on its behalf in any country . . .'. On 9 August, Havlin applied for a visa at the Paris office of Intourist. Not only was Rome never mentioned in his eight futile visits to Intourist and to the Soviet Embassy, but he was repeatedly assured that the Soviet Embassy in Paris would issue the visa imminently. This was the case until 9 September when an official in the Embassy told Havlin that there was no chance that he would receive a visa. These facts should be contrasted with the

picture presented by Zheludev.

Second, Zheludev raised the possibility that "... we could judge the fact that just two scientists tried to participate instead of the four we were informed of, as an additional difficulty created intentionally." Besides ourselves, Dr Lucien Benguigui of the Technion and Dr Yosef Yeshurun of Bar-Ilan University wrote numerous times to the Organising Committee requesting information regarding attendance at the meeting. When they received no replies, they abandoned further attempts to attend. To imply that we were attempting to "create difficulties" by having these individuals express interest in attendance and then failing to appear in Rome or Vienna to collect visas for which they had not applied or been informed of, is preposterous.

Third, Zheludev complained that we publicised the incident before clarifying details. We have already mentioned that every one of our many letters to the organising committee seeking to clarify matters went unanswered (with the sole exception of the cable to IUPAP). Only after our letter was submitted to *Nature*, with copies to several international bodies, did the organising committee see fit to respond, both with Zheludev's letter to *Nature* and a copy sent directly to us in Israel by normal mail service.

In conclusion, we re-emphasise the profound need for the recommendations which we proposed in our letter of 16 February. The issue is simple: hosting an international conference means fully accepting the responsibility for insuring the possibility of attendance by scientists from all countries. This right must be strictly guaranteed for Israeli scientists as it should be guaranteed for scientists from all other countries.

Yours faithfully,

SIDNEY B. LANG

*Ben Gurion University of the Negev
Beer Sheva, Israel*

SHLOMO HAVLIN

*Bar-Ilan University
Ramat Gan, Israel*

SIR,—I would like to let you know my position concerning the Cancer Congress in Buenos Aires.

Whilst there are certain people who will go to this congress without restriction, there are others who propose to boycott it and will do so.

I wrote to the Chief of State of Argentina requesting an audience in order to present him with the list of those scientists of whom we have no news, making this audience the condition for my coming to Buenos Aires.

Since this audience was refused, I wish to let you know that I will not go to Argentina. I consider that this flat refusal gives weight to the argument of those people planning to boycott the meeting.

Yours faithfully,

GEORGES MATHÉ

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news and views

Holes in the microwave background?

from M. Birkinshaw and S. F. Gull

RADIO sources lying outside prominent clusters of galaxies typically appear as two bright lobes of emission on either side of an 'active' galaxy. Those sources within the great clusters, on the other hand, often have highly irregular shapes. One hypothesis advanced to explain this phenomenon is that each cluster of galaxies holds an atmosphere of very hot (10^7 – 10^8 K) gas which affects the evolution of any radio source within it. The discovery of cluster X-ray sources seemed to confirm the existence of these atmospheres—X-radiation being the predominant thermal bremsstrahlung of gases at such temperatures. A strong test of this model is provided by observations of the temperature of the microwave background (the thermal radiation surviving from the 'big-bang' and now at about 2.9 K) towards an X-ray cluster; the thermal model predicting that it should be depressed by about 10^{-3} K due to the presence of hot gas. No such depression is expected from the main rival model—indeed an excess of radio sources, giving a temperature enhancement, would be anticipated.

Some of the problems associated with the very difficult observations needed to find a temperature decrement of 10^{-3} K have been highlighted recently by J. Tarter (*Astrophys. J.* **220**, 749; 1978). The experiment is performed by continuously measuring the temperature difference between directions towards the centre of a test cluster and reference points outside it. This method removes most of the effect of our atmosphere and saves having to measure temperatures of 2.899 and 2.900 K independently to high accuracy. However, since the expected

cooling is very small, it would be swamped by even the most modest radio source lying along the line to the cluster, or would be falsely produced by that source lying near the reference points—so both regions have to be checked carefully for interfering sources. The effect of radio sources is diminished by working at high frequencies, but here the atmosphere becomes unstable, and temperature fluctuations of some hundreds of 10^{-3} K may occur. This forces the use of a non-ideal frequency (10.6 and 33 GHz have been used), and the main and reference beams of the telescope have to be set close together. Unfortunately both beams then lie within the gas distribution of the test cluster—so less temperature decrement is expected and the experiment is made harder.

Even then there are problems. If any cooler gas (at 10^4 – 10^6 K) lies within the cluster, it will emit radio radiation in the same way as the hotter gas emits X-radiation. Only a little cool gas is needed to swamp the expected temperature decrement. Last, but not least, is the problem of observing time. Long-term atmospheric instabilities are such that about 60 h of observing at any one point is necessary to establish the presence or absence of a temperature decrement at a useful level, and telescope time is more commonly allocated by the night than by the week or month.

Despite these difficulties a number of papers reporting detections or limits to the magnitude of the background cooling have now appeared (for example, Parijskij *IAU Symp.* 79; Gull & Northover *Nature* **263**, 572; 1976; Lake & Partridge *Nature* **270**, 502; 1977). The disagreements among these observations can, in general, be explained by differences in the precise experimental details. Four out of a total of 11 clusters observed have been

found to show the background cooling—though one of these (Abell 2125) is a subject of some debate. Two of the clusters have now been scanned to show the extent of their atmospheres. Furthermore, on the basis of work on the background radiation, the cluster Abell 2218 was predicted to be an X-ray source. This was later confirmed by observations made on the Ariel V satellite. These results indicate strongly that the diffuse cluster X-ray emission has a thermal origin—and allow measurements of the density of the emitting gas. The detection of the decrement also constitutes proof that the microwave background really is of cosmological origin and exists on scales of at least 3,500 million light-years.

Microwave background astronomy has tended, in the past, to concentrate on the measurement of the radiation's spectrum—to confirm its thermal nature. Early attempts to search for slight anisotropies in the background due to the peculiar velocity of the Earth or the presence of condensing clusters of galaxies (protoclusters) were severely limited by the available technology; but recent improvements in telescopes and receivers have made high-precision measurements possible—as shown by the detection of the motion of the Earth relative to the background radiation by Smoot *et al.* (*Phys. Rev. Lett.* **39**, 898; 1977). Although protoclusters have not yet been detected, further work should reveal them and provide important information on the evolution of structure in the present-day Universe. The new generation of X-ray telescopes and microwave receivers will undoubtedly provoke more work on the structure of gas distributions within clusters—and with this superior information on the environment of radio sources some of their cryptography may, at last, be decoded. □

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Evolutionary conservation of gene linkage

from E. H. R. Ford

THE science of chromosome mapping in mammals has grown explosively in the past 10 years, largely because of the development of the techniques of chromosome banding and of somatic cell hybridisation. In man, over 110 gene loci have been assigned to specific autosomes, and 100 more to the X chromosome (McKusick & Ruddle *Science* **196**, 390; 1977). This is the highest number of gene assignments for any mammal, and the next highest number is in the mouse. In both man and mouse a number of gene linkage groups have been established by family and breeding studies, and it is now possible to show that some of these have been conserved in both (and other) species during the course of evolution.

The technique of somatic cell hybridisation depends on the fusion of cells from two different species, following which the chromosomes of one of the two species 'segregate' (are preferentially removed from the hybrid cell). Normally if human and rodent cells are fused, the human chromosomes are discarded, and this has been invaluable in constructing the human chromosome map (since the gene loci for enzymes can be located by studying which human chromosomes remain after fusion with rodent cells deficient in that enzyme). However J. D. Minna and his co-workers discovered that if cultured human cells are fused with differentiated rodent cells, the rodent cells segregate, and this discovery has been as valuable in building up the mouse chromosome map as the original one was for the human map. Somatic cell hybridisation is also valuable for showing which genes are 'syntenic', that is, located on the same chromosome, whether or not they are linked (Renwick *A. Rev. Genet.* **5**, 81; 1971). It does not however tell whether genes are linked, and if so how closely; this information must still be obtained by sexual genetics.

Now Lalley, Minna and Francke (page 160 of this issue of *Nature*; see also *Proc. natn. Acad. Sci. U.S.A.* **75**, 2382; 1978), inspired by the recent finding that five mouse genes are syntenic, and that these are also syntenic in chromosome 1 in man (four of them being assigned to the short arm), have sought other examples of genes which are syntenic in both mouse and man. Following their assignment of seven more gene loci to specific mouse chromosomes by somatic cell hybridisation, they have

found evidence for synteny of four pairs of gene loci on four different human and mouse autosomes. When these findings are combined with previous ones by this and other groups, it seems that there is now evidence for nine pairs or groups of genes which are syntenic on different autosomes in both man and mouse, as well as a cluster of genes which is syntenic on the X chromosome in both species.

It has been known for some time that some groups of very closely linked genes are conserved together (and are therefore of course syntenic) in several species—for example the major histocompatibility complex in man, rhesus monkey and mouse, the four γ -immunoglobulin heavy chain genes in man, mouse and rabbit, pancreatic and salivary amylase genes (man, mouse, *Drosophila*) and the β and γ polypeptide chains of haemoglobin (man and mouse). A plausible explanation of this is that the genes concerned are closely related to each other both in structure and function, and may indeed have arisen during evolution by tandem repeats (Ohno *Nature* **244**, 259; 1973). There is every reason to suppose that such groups would be conserved, both because they are so very closely linked and because they are functionally interrelated. But the new findings of Lalley *et al.* suggest that quite distantly linked genes may be kept together on the same chromosome; for example the genes for phosphoglucomutase-2 and phosphogluconate dehydrogenase are 23 map units apart in the mouse and not detectably linked in man.

The evolutionary significance of the preservation or disruption of syntenic groups of genes is, as Lalley *et al.* say, not yet understood. Obviously during mammalian evolution much chopping and changing of chromosome segments has occurred (from reciprocal translocations and inversions for example). In closely-related species such as man and chimpanzee these can be analysed and largely explained, but when species are as distantly related as man and mouse, it is generally difficult if not impossible to make homologies between the chromosome banding patterns or to reconstruct the chromosomal changes which have taken place in the course of evolution. There is however one notable exception to this, which is the mammalian X chromosome; this shows similar banding in some quite distantly related species (for example man and horse). Here Ohno's law, which suggests that if genes are found to be present or linked on the X chromosome of one

mammalian species, they may similarly be expected also to be present or linked on the X chromosomes of any other species appears on present evidence to have considerable (if not total) validity.

It may be that, as Lalley *et al.* suggest, chromosomal rearrangements which tend to preserve ancestral synteny are preferentially favoured in evolution. One obvious example of this is the Robertsonian translocation (centric fusion) whereby two acrocentric chromosomes combine to form a single metacentric. This is widely documented as one of the commonest evolutionary changes both in primates and in other mammalian groups, and it is noteworthy that it does not involve any rearrangement of linkage groups.

The development of gene maps in different species, which is now progressing so vigorously, is clearly going to add profoundly to our understanding of the mammalian genome, and of the underlying mechanisms of its evolution, as well as to our knowledge of the functional relationships between different genes and clusters of genes. □

Large-scale characteristics of the Galaxy

from Victor Clube

NEARLY two hundred astronomers gathered recently at the University of Maryland for the 84th Symposium of the International Astronomical Union, entitled 'The Large-Scale Characteristics of The Galaxy'. The aim of the gathering was to service and tune up The Working Model of our Galaxy. The machinery had evidently sprung a good many leaks, and though this was not allowed to be a cause for too much public concern, all is not well.

The Model is basically of 1920 vintage though it was called in for a major overhaul in the 1950s. It is now showing signs of obsolescence. The Model is at heart a spheroidal distribution of stars circulating interminably in epicyclic orbits. By the 1950s, developments in the theory of stellar evolution and the advance of radio astronomy had also required a

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role for gas and dust. An essentially never ending cycle was therefore set up in which the gas shed by evolving stars was fed into a rotating disk to be then pressed by spiral density waves into making just enough new stars to replenish the general stock. For a while, the machine took on a new lease of life and got quite good at explaining the chemical content of the Galaxy. But now in the 1970s, we are again in trouble—new diagnostic tools such as the infrared and millimetre probes have shown that the machine is not functioning at all correctly. When, too, we look out at other galaxies and see so many of them as 'peculiar', the suspicion can arise that we may be missing something of fundamental importance. Perhaps we should keep in mind the proverbial Tibetan monks who, having watched a cricket match on a none too good television set, were driven to write to the MCC for all the equipment, but omitted to ask for a ball?

J. H. Oort (Leiden) summarising conditions at the galactic centre came nearest to the problem when admitting the role played by the nucleus was simply not known. The limits of our knowledge thus defined, no one dared address the rather basic question whether the nucleus is a 'minor perturbation' or a 'large-scale characteristic'. As usual, when confronted with the unknown, there are always those who will assure us everything is normal and under control. After all, there is a dearth of X rays and the nearby Ne II clouds are none too sprightly. Even if there is a black hole, we are told, it cannot be very massive—so, it is assuredly of no consequence. Others too, could only see rather ordinary star formation processes at play around the nucleus while the infrared seems to be mostly that of normal stars and dust. Someone does remember the awkward velocity of Sgr A West and tries to shift the galactic centre slightly to the right, but the chairman quickly moves us on. Then, we hear an impressively tidy description by W. B. Burton (NRAO) of the molecular clouds hereabouts in terms of an off-axis rotating, expanding disk. Everything does seem to be under control. But—how does the disk work? In come the heavyweights—no problem: if you think hard enough, you can convince yourself the expansion is not there—it's all done with bars. Can anyone see a bar? No? Oh well, never mind—there are lots of bars in other galaxies. Now we are safe—the solution is beyond reach!

The density wavers quickly come and go, rather exasperated to find very little in the Galaxy to favour their invention. It seems from looking at other galaxies, it might help to be a long way off to

get this theory to work also! Can the density waves really produce all those massive CO clouds in the 5 kpc ring? and why don't they show spiral structure? After all, the HI and CO in the galactic centre and regions of star formation in the solar vicinity always seem to go together. And how do we avoid them making too many stars? Not to put too fine a point on things, it is as if the CO clouds appear from nowhere in a distribution we don't understand and turn into stars by an unknown process! There are evidently big mysteries here, but at least it was agreed that southern hemisphere observations would certainly help. The ultimate solution of these mysteries obviously invites a good deal of speculation and the idea that supernova-driven shocks are the principal cause of star formation finds favour in some quarters. The very wide range of physical conditions encountered in the gaseous disk was emphasised by E. E. Salpeter (Cornell University), but they are not readily explained by the supernova model. Indeed the cloud structure evidently has more the character of 'sheets'. P. L. Baker (Max-Planck-Institut) saw these as transient features which may be related to star streams. He seemed to be wondering whether we were suffering a patch of bad 'galactic weather'. The sceptic might wonder if all this evidence does not indeed point to some major instability on the galactic scale.

Although there is no great desire to alter the scale of the Galaxy much away from $R_0 = 9$ kpc, the kinematic picture in the outer regions of the Galaxy is now extremely confused. If M-stars observations by Uppgren (van Vleck Observatory) are to be believed, some non-circular motions can hardly be avoided, but many would prefer not to notice, hoping the problem will go away. The standard circular velocity in the solar neighbourhood of 250 km s^{-1} will almost certainly survive however for the time being since those advocating higher and lower values at this symposium were just about evenly balanced! V. Rubin (Carnegie Institute) showed beautiful flat rotation curves for many galaxies and this was seized upon as evidence for the same in our own. Gravitational test particles in the outer reaches of the Galaxy seem also to be dancing the right tune for a 'massive halo' according to J. Einasto (Estonia), and the assembled astronomers were happy to see the nearest 'missing matter' coming within reach in the Galaxy. In the euphoria, it is forgotten we still cannot see it, and perhaps it's not so good for 'warps' either. As well as warps, there are the enigmatical high velocity clouds also observed out of the galactic plane. G. L. Verschuur (Colorado) emphasised

the likelihood of a variety of origins, and without question, many of them are associated with the so-called Magellanic Stream. At the same time, many are not, and R. Giovanelli (Oklahoma) considered it a sign of weakness not to attempt a unified explanation. None was forthcoming, but in his search, he does seem to have found a previously unknown dwarf spiral. Admirable though this is, it does perhaps illustrate the principal problem of this symposium—how to tell the wood from the trees. Without a doubt, it has not been the last. □

Anaerobes and transferable drug resistance

from J. R. Saunders

THE obligately anaerobic bacteria of the genus *Bacteroides* are normal inhabitants of the gut and oral cavities of man and other animals. Until recently these organisms were rarely implicated as causative agents of human disease. However, advances in the culturing and identification of *Bacteroides* species have revealed that such anaerobes are involved in a significant number of clinical conditions including wound infections and septicemia. The evolution of drug resistance in *Bacteroides* is therefore of some importance.

In this week's *Nature* (page 181) Donald Guiney and Charles Davis report the identification of a conjugative resistance plasmid present in a strain of *Bacteroides ochraceus* which is a resident of the oral cavity. This R plasmid specifies resistance to chloramphenicol, tetracycline and kanamycin and is transmissible from *B. ochraceus* to *Escherichia coli*. The presence of an R factor mediating resistance to kanamycin in *Bacteroides* is in itself rather surprising, as anaerobes are usually insusceptible to such aminoglycoside antibiotics (see *News & Views* 8, 475; 1977). There have also been recent reports (see, for example, Mancini & Behme *J. Infect. Dis.* 136, 597; 1977) of self-transmissible R plasmids in *B. fragilis*. This organism is a major constituent of the normal flora of the large intestine and is generally the principal cause of infections due to anaerobes.

The finding of R plasmids with sex factor activity in *Bacteroides* has a number of important implications. *Bacteroides*, in particular *B. fragilis*, present a number of problems in

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chemotherapy, as they are relatively resistant to a number of antibiotics. Some strains of *B. fragilis* are particularly resistant to β -lactam antibiotics (penicillins and cephalosporins) because they possess a β -lactamase (see, for example, Britz & Wilkinson *Antimicrob. Ag. Chemother.* **13**, 373; 1978) which is active even against some of the new generation of ' β -lactamase-resistant' cephalosporins. Fortunately, the genetic determinants of this β -lactamase seem to be chromosomally located, reducing the immediate possibility that they will spread to other *Bacteroides* species which are currently fairly sensitive to β -lactams. However, the presence of conjugative R plasmids in this genus must inevitably hasten the wholesale acquisition of resistance to these and other antibiotics.

In the colon, obligate anaerobes outnumber facultative anaerobes such as *E. coli* by as much as 1,000 to 10,000-fold. Such large populations may therefore provide considerable scope for the evolution of new R plasmids. The R plasmid described by Guiney and Davis in *B. ohraceus* and those of *B. fragilis* are capable of transfer to *E. coli*. These obligate anaerobes could therefore provide an extensive pool of resistance plasmids capable of infecting more commonly recognised pathogens such as *Salmonella* and *Shigella*. It is not yet clear whether *Bacteroides* R plasmids originate within this genus or whether they have been acquired from other bacteria. It is known that *Bacteroides* spp. can receive a number of R plasmids from *E. coli* provided that recipient strains are heated to 50°C for 20 min immediately before mating, a procedure which presumably inactivates their restriction systems (Burt & Woods *J. gen. Microbiol.* **93**, 405; 1976). A more detailed examination of *Bacteroides* plasmids by incompatibility testing and DNA sequence homologies should reveal whether they correspond to types of plasmid already known, in for example the Enterobacteriaceae.

It is worth noting that in laboratory experiments *Bacteroides*, if present in the high concentration found in the human intestine, are claimed to inhibit transfer of R plasmids between strains of *E. coli* (Anderson *J. med. Microbiol.* **8**, 83; 1975). This inhibition of transfer does not seem to be due to mechanical interference but is probably due to a metabolite produced by *Bacteroides*. Whether or not this reflects the situation in the human gut has not been ascertained. Other factors, such as the presence of bile salts and anaerobiosis *per se*, may affect transfer frequencies between donor and recipient strains of *E. coli* *in vivo*. In strictly anaerobic conditions

in vitro R plasmids of some incompatibility groups do show reduced rates of transfer, whereas others show little effect (Burman *J. Bact.* **131**, 69; 1977). Certainly an anaerobic environment provides no absolute barrier to bacterial conjugation, as witnessed both by the self-transmissible R plasmids of *Bacteroides* and those recently described in *Clostridium perfringens* (Brefort *et al. Plasmid* **1**, 52; 1977).

The emergence of transferable drug resistance in *Bacteroides* is yet another example of bacterial populations responding to the selective pressure imposed by widespread use of antibiotics. This will undoubtedly increase the clinical problems associated with treating conditions caused by these organisms. As major human and animal commensals *Bacteroides* harbouring resistance genes also provide a potential threat to health by transmitting these determinants to more conventional pathogens. □

Extragalactic molecular hydrogen

from C. G. Wynn-Williams

A GROUP of infrared astronomers using the 2.3 m telescope at the Steward Observatory, Arizona, have recently announced the first direct detection of molecular hydrogen in a galaxy outside our own Milky Way (Thompson, Lebofsky & Rieke *Astrophys. J.* **222**, L49; 1978). The surprising feature of this discovery is that the galaxy in which the H₂ was found is not one of our near neighbours such as the Andromeda Nebula, but is NGC 1068, a well-known Seyfert Galaxy. At first sight this result is paradoxical. Interstellar molecules, as opposed to the more widespread free atoms and ions, exist mainly in dense, cold slow-moving clouds; the nuclei of Seyfert Galaxies, on the other hand, are renowned for their violent motions and their powerful ultraviolet radiation fields. The fact that molecules can survive under these conditions provides an important clue to the processes occurring in galactic nuclei and QSOs.

Although molecular hydrogen is much the most abundant of the forty or so molecules so far detected in interstellar space, it is one of the most difficult to observe since it has no spectral lines at radio or millimetre wavelengths. The rapid growth of molecular-line astronomy during the past decade has been based to a large extent on observations of transitions between adjacent rotational levels of the ground

states of simple organic molecules. In dense interstellar clouds, such as those in the process of condensing to form new stars, the upper rotational levels are excited as a result of thermal collisions within the gas which has, typically, kinetic temperature of 50 K. Molecules which have permanent dipole moments, such as CO and HCN, can then decay spontaneously to the lower state, giving rise to the spectral lines studied by radioastronomers. For symmetric molecules such as H₂, however, these transitions are forbidden and unobservable. The famous 21-cm spectral line of atomic hydrogen is also absent from the dimer. The presence of H₂ molecules can often be inferred indirectly by millimetre-wave observation of carbon monoxide and other molecules, but to study molecular hydrogen directly it is necessary to move to other less accessible wavelength ranges.

Rocket and satellite-borne telescopes have been used to study the ultraviolet spectrum of H₂ in absorption in front of bright stars, but problems of sensitivity and interstellar absorption by dust grains have confined these observations to relatively transparent low-density clouds close to the sun. The first successful infrared detection of molecular hydrogen was announced in 1976 when a group at the University of Arizona identified seven emission lines in the spectrum of the cloud OMC1, behind the Orion Nebula (Gautier *et al. Astrophys. J.* **207**, L129; 1976). These lines are rotation-vibration quadrupole transitions between the ground and first vibrationally excited states of H₂; they lie in the wavelength range 2.0 to 2.5 μ m, where the Earth's atmosphere is relatively transparent. The same lines, or at least some of them, were subsequently detected in six other Galactic objects (Treffers *et al. Astrophys. J.* **209**, 793; 1976, Beckwith *et al. Astrophys. J.* **219**, L33; 1978). Unlike the Orion cloud, which is a region of active star formation, these latter sources are all planetary nebulae, small clouds of material ejected from the surface of old stars.

What has puzzled astronomers about the infrared detection of H₂, particularly in the Orion source, is the method by which the upper vibrational level is excited. If the excitation were thermal, a gas temperature of 2,000 K would be needed; such a temperature is at least an order of magnitude higher than that determined for the cloud from other fairly reliable evidence. A more specialised process is therefore necessary. The mechanism currently favoured is that of shock waves within the dense regions of the cloud, moving through it at such velocity, 10 to 20 km s⁻¹, that they are strong enough to provide the energy to excite the $v = 1$ transition, but not so strong as to dissociate the

molecules completely (Hollenbach & Shull *Astrophys. J.* **216**, 419; 1977; London *et al. Astrophys. J.* **217**, 442; 1977). The origin of these shock waves is uncertain. None of the known stars or protostars in the Orion region is powerful enough to maintain these shocks; the possibility that a cataclysmic event such as a supernova has taken place deep within the Orion cloud is therefore being seriously considered (Kwan *Astrophys. J.* **216**, 713; 1977). Indirect support for the notion that something unusual has occurred in the Orion cloud comes from the fact that searches for similar H_2 emission from other regions of star formation in our Galaxy have so far been unsuccessful.

It is this possible link between dense gas clouds and explosive events which makes the discovery of infrared H_2 emission from NGC 1068 of great interest. NGC 1068 is one of the nearest 'Seyfert' galaxies. These objects, of which about 100 are now known, are characterised by their very luminous nuclei and by the strong emission lines they display at visible wavelengths. These emission lines arise from clouds of hot ionised, as opposed to molecular, gas, and are very broad in profile, with Doppler velocity widths of the order of $1,000 \text{ km s}^{-1}$. The Seyfert nuclei also show synchrotron emission at radio wavelengths. The observed properties of Seyfert galaxy nuclei suggest that the processes occurring there are related to those in radio galaxies and quasars, albeit on a smaller scale. One important property of Seyfert galaxies is that they are very powerful in the wavelength range 5 to $200 \mu\text{m}$. In fact, the infrared emission from the nucleus frequently exceeds that from the rest of the galaxy at all other wavelengths. In most cases, perhaps all, the bulk of the infrared emission originates from microscopic dust grains at temperatures in the range 30–300 K. At the moment we do not know whether these dust grains are spread uniformly throughout the nuclear region, as in our Galaxy, or concentrated into dense clouds such as those which herald star formation. The discovery of molecular hydrogen in NGC 1068 seems to favour the latter model, although the possibility remains that the H_2 emission comes from a large number of old objects such as planetary nebulae. In either case it is significant that the Doppler width of the molecular hydrogen is comparable to that of the atomic species, indicating that the clouds themselves are involved in the violent motions of the nuclear regions.

Further observations of molecular hydrogen in Seyfert and other related galaxies should help us to establish how much of the violent activity in these

objects is attributable to 'ordinary' processes such as bursts of star formation, and how much of it depends on the existence of an exotic compact object within the nucleus. □

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Clocks, pineals and compasses

from J. Krebs

THE fact that living organisms have internal clocks is well established: a large number of physiological and behavioural rhythms continue their daily (circadian) oscillations under strictly controlled constant environmental conditions. One of the major goals of current work on circadian clocks is to identify their physiological basis, and small birds have proved to be well designed for tackling this problem because they have striking circadian activity rhythms, which can be recorded automatically in a cage by perches equipped with microswitches. At the XVIIth International Ornithological Congress*, M. Menaker (University of Texas, Austin) summarised a series of exquisitely ingenious experiments with house sparrows, the sum of which show that birds may have circadian master clocks located in the pineal gland, a tiny light-sensitive neurosecretory structure in the brain and thought to have functioned as an eye in some ancestral vertebrates.

When the pineal is removed from sparrows kept in constant darkness, the normal endogenous (free running) rhythm of activity breaks down: instead of showing a clearcut daily alternation of active and inactive periods, the birds become arrhythmic. The pineal has both neural and vascular connections with the rest of the brain, but by severing the nerves, Menaker's group were able to show that the pineal's influence on circadian clocks is transmitted by way of the bloodstream. A chemical, rather than neural link is also indicated by experiments in which Menaker and his coworkers restored the endogenous activity rhythm to pinealectomised sparrows by pineal transplants into the anterior chamber of the eye. Here the tissue thrives and continues to secrete chemical messages, but presumably does not initially form any neural connections with the recipient. These results show

that the pineal in birds (though it must be stressed that the same does not seem to hold for mammals) has a central role in circadian activity rhythms. They do not, however, say whether the pineal generates the rhythms or whether it acts only as an intermediary, transmitting messages from clocks located elsewhere in the body. Menaker's most persuasive evidence that the pineal actually generates rhythms is the so-called phase transplant experiment. In this, two groups of sparrows whose activity rhythms are entrained by a dark-light cycle to be 180° out of phase with one another are used as pineal donors to two groups of arrhythmic, pinealectomised, recipients living in constant darkness. The remarkable result of this experiment is that the recipient bird adopts the phase of the donor's pineal. Two birds receiving pineals from two donors 180° out of phase will initiate their new free running rhythms with the same 180° phase difference. It seems therefore as if the avian pineal could be the master clock controlling activity and perhaps other circadian rhythms.

One of the hormones produced in the pineal is melatonin, and at least three lines of evidence suggest that melatonin is involved in circadian rhythmicity: the pineal has a rhythmic output of a melatonin precursor which is maintained *in vitro*; melatonin implants (giving a continuous output) can lead to a breakdown of the circadian activity rhythms in sparrows, and finally daily pulses of melatonin injected into pinealectomised starlings can act as a synchroniser for the activity rhythm. This last result was reported by E. Gwinner (Max-Planck-Institut, Erling Andechs), who also reported that the activity of pinealectomised starlings does not become completely arrhythmic as in sparrows, although the pineal certainly has a drastic influence on the rhythm. This leaves open the question of whether different species of birds differ in the extent of pineal control, or whether there may be after all some subordinate self-sustained oscillators which are synchronised by the pineal.

One of the ways in which birds can use internal clocks is, in combination with the Sun, as part of their extensive armoury of compass mechanisms used in migration and homing. The most recent work on navigation and compass orientation in birds has concentrated on scent and magnetism. In addition to several symposia devoted to navigation, W. T. Keeton (Cornell University) reviewed the whole field in his usual eloquent and lucid style. Among new findings since I last reported on the subject in 1975 (*News & Views* **257**, 358) are the following: magnetic information on the outward journey seems

*The XVIIth International Ornithological Congress was held in West Berlin, on 4–11 June.

to be important in pigeon homing (shown by F. Papi's group at Pisa, Italy, who transported pigeons to their release site in steel boxes). Pigeons in the laboratory have been found to be extraordinarily sensitive to infrasound, barometric pressure, ultraviolet light and polarised light (M. Kreithin, Cornell University) all of which could be used as cues in homing. The delightful idea that pigeons smell their way home

(F. Papi and coworkers) is still in dispute, the Cornell group being unable to show the deleterious effect of various scent manipulations on initial orientation of pigeons after release shown so convincingly by the Italians. In one experiment the two groups had collaborated and found that interfering with the bird's sense of smell did not influence initial orientation but decreased homing success, and the results were interpreted in different ways by the two groups, emphasising that even hard data have to undergo subjective interpretation. □

gallium arsenide cell (which is suitable for concentrated sunlight up to several thousand Suns) again comes into its own and its efficiency of order 20% may be expected to be raised to 30% if it is linked to a germanium cell, and this might be increased to 35% for a three-cell system at one Sun. A rather academic 10 cell system might yield 50%. The only experimental results so far available refer, however, not to a single unit containing several cells but to physically separate cells, receiving separately radiation in their spectral range. Here an efficiency of 28% at a concentration of 165 Suns has been attained by Varian Associates. The spectral division is accomplished by a device (a 'dichroic mirror') which transmits with minimal loss (of order 5%) the spectral range which it does not reflect.

The cost reduction of silicon solar cells is kept firmly in view as 1986 approaches, when the US Department of Energy aims to be producing electricity from solar cells at the rate of \$0.50 per peak watt. Cheaper ways of producing silicon are therefore required. The Union Carbide Corporation has developed a new three-step, low-cost process which yields p-type silicon, without the need to add impurities, starting from metallurgical silicon. After phosphorus diffusion to make a p-n junction this material yields 8% efficient solar cells. The cells are therefore comparable with polycrystalline cells made from higher (semiconductor) grade silicon, which themselves represented one of the advances reported at the last Photovoltaic Specialist Conference. This is therefore certainly a step in the direction of lower costs.

New advances were also reported on methods of concentrating radiation. The concept of a luminescent solar concentrator has been taken further by Owens-Illinois, Inc. It provides concentration of sunlight without the need for a mechanism to track the Sun in its apparent motion, and it also manages to concentrate radiation. This is an important component of solar radiation, particularly on cloudy days, and is difficult to concentrate by mirrors or lenses. In the new device the radiation is absorbed by a fluorescent sheet, and re-emitted as luminescence. This cannot escape because of internal reflection at the plane surfaces of the sheet and because the edges are silvered. The trapped radiation is then incident on a photocell. Improvements in appropriate absorbing dyes and in other ways are still needed. Another method of developing concentrators of direct

Progress in solar cells

from P. T. Landsberg

THE energy crisis has stimulated increased research and development on solar cells, which convert solar radiation directly into energy. Advances in this field were reported recently at the 13th Photovoltaic Specialist Conference.*

The chief use for solar cells at present is in providing power for satellites. These solar cells, must be light, with good end-of-life efficiency. The 1973-silicon cell for space, 350 μm thick, has now been replaced by a 50- μm cell which uses a back-surface field and is more resistant to the electron and proton bombardment to which it is subjected in space. Unless their open-circuit voltage can be improved, J. Scott-Monck (Jet Propulsion Laboratory, Pasadena) did not foresee much improvement over the present efficiency which depends on the radiation level, type of cell, and so on, but is typically 15% at room temperatures. For the future it is important to reduce remaining traces of oxygen, boron, carbon, and possibly lithium and to improve the radiation resistance. The advance by a factor of 7:1 in the power/weight ratio is largely due to contracts awarded some years ago, and now completed, to Solarex Corporation, Spectrolab Inc., and Optical Coating Laboratory Inc., firms deeply involved in the solar cell business. The claims of gallium arsenide cells will also have to be considered more seriously for space cells. However, they need to be made lighter and more reliable. As regards the remaining four of the six criteria for space cells—availability of the raw material, cost, efficiency and radiation resistance—gallium arsenide seems about as satisfactory as silicon cells.

The possibility of using gallium arsenide in space has made it essential to study the properties of these cells under electron and proton bombardment. Since most damage is created at the end of the travel of the incoming particles one would expect to require a sensitive region in the solar cell which is thin enough. Since long wavelength photons are absorbed less rapidly than short wave ones, degradation due to bombardment should show up most markedly in that part of the electric power output which is the result of conversion of the longer wavelength photons. These expectations have now been better quantified: a limit of order 2 μm has been put on the thickness of the sensitive region; the worst damage has been found in the spectral response wavelengths in excess of 0.75 μm ; both numbers apply for certain typical conditions. In addition damage coefficients have now been determined. In terrestrial silicon solar cells at least one type of degradation was suggested by H. Brandhorst (NASA-Lewis Research Center, Cleveland) to be due to silver impurity atoms, which may diffuse from the contact materials.

In the terrestrial field, the idea of providing separate solar cells for different parts of the solar spectrum, although not new, has now attracted a number of feasibility studies. The idea is to split the solar spectrum into ranges, and to optimise a cell for the range which it is supposed to convert. Cost, complication of structure, and the second law of thermodynamics would then be eventually the only limits on the efficiency. In order to bring the cost per watt of a unit down again, these 'tandem cells' are being considered largely in conjunction with means of concentrating the solar radiation falling on the unit. The

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*Held on 5-8 June, 1978 in Washington, DC.

radiation is to make them less dependent on accurate tracking, and such designs were also reported.

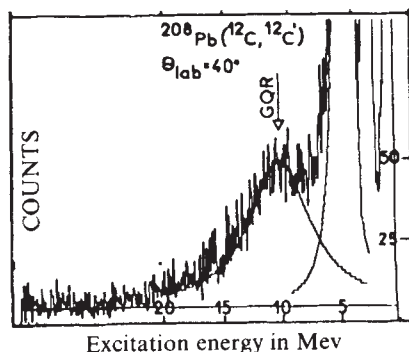
The meeting showed clearly that the photovoltaic community in the United States is rapidly growing. The United States administration is clearly determined to make a success of photovoltaics. Indeed the considerable sum already being channelled by the US Department of Energy into this work is liable to be further increased if a Bill introduced by Congressman Mike McCormack (Democrat, Washington) is approved. This 'Solar Photovoltaic Energy Research Development and Demonstration Act of 1978' calls for the expenditure of an additional 1.5 thousand million dollars over the next ten years. This would give 125 million dollars for the fiscal year 1979. With expenditure of this magnitude many more scientists, technologists and industrial firms must be expected to apply their ingenuity to these problems. □

Excitation of nuclear giant resonances by heavy ions

from P. E. Hodgson

THERE is currently great interest in the collective modes of oscillation of highly excited nuclei. In these collective excitations, the nucleons move together in a correlated way, in contrast to the single-particle excitations which involve only one or a very few nucleons. The simplest examples of collective excitation are the vibrations of the soft nuclei and the rotations of the deformed hard nuclei, and in addition there are more energetic excitations that involve the bulk motion of the protons relative to the neutrons.

Such modes of oscillation can be excited by a projectile that exerts a strong electric field; this acts on the protons but not on the neutrons, and so the protons are all pulled away from the neutrons, and thereafter the protons and neutrons oscillate relative to each other. These excitations produce a broad maximum in the cross section of the reaction that excites them, and these are known as giant resonances. The first giant resonance to be studied was the dipole resonance at about 22 MeV in light nuclei, but since then many higher resonances have been found using a variety of projectiles and reactions. As might be expected from their collective character, the



Energy spectrum of ^{12}C ions inelastically scattered by ^{208}Pb showing the peak due to the excitation of the giant quadrupole resonance.

energies and widths of the resonances change rather slowly from one nucleus to the next, as they are not dependent on the finer details of nuclear structure.

The first theories of the giant resonances used the hydrodynamical model of two fluids oscillating relative to each other, and this was able to account for the slow variation of the energy with mass number. Later on, microscopic theories were developed, and these attribute the excitation to a coherent superposition of all possible particle-hole excitations of the nucleons from one shell to the next. The theory is being developed and applied to the higher resonances. In particular, it has been found that they are strongly excited by proton inelastic scattering, and the cross-sections have been analysed to provide evidence for the quadrupole ($L=2$), octupole ($L=3$) and hexadecapole ($L=4$) resonances in addition to the familiar dipole resonance (see *Nature* **246**, 250; 1973). Other studies have been made using deuterons, helions and α -particles as bombarding particles.

This work has recently been extended by Buenerd *et al.* of Grenoble using the heavy ions ^{12}C and ^{14}N as projectiles to excite giant resonances in a series of nuclei from ^{40}Ca to ^{209}Bi (*Phys. Rev. Lett.* **40**, 1482; 1978). Heavy ion reactions have the advantage that they can transfer a large amount of orbital angular momentum to the target nucleus, and thus excite resonances that cannot be excited by lighter projectiles. It is also to be expected that some competing processes such as quasi-free and pre-compound emission are unlikely in heavy ion reactions, so that the background is less and the giant resonances are correspondingly more prominent.

In the new experiments, each nucleus was studied by measuring the energy distribution of the inelastically scattered ions near the grazing angle, to maximise the probability of high momentum transfer. One of these

spectra is shown in the figure, and it is apparent that the peak due to the giant resonance is clearly resolved. In the case of the resonance excited by ^{14}N on ^{208}Pb the cross section was measured at several angles and a distorted wave calculation with $L=2$ and a deformation parameter of 0.09 was found to fit the data well, thus confirming the assignment to the giant quadrupole resonance. The integrated cross section corresponds to about 90% of the energy-weighted sum-rule limit for E2 transitions, and agrees with the results of other determinations.

These experiments show that heavy ions can excite the giant resonances in a wide range of nuclei with good intensity relative to the background. Comparison with the results obtained using lighter projectiles sometimes show differences, and these could be due to the presence of higher multipolarities in the heavy ion reactions, and further work could lead to their identification and analysis. This work establishes heavy ions as powerful probes of highly-excited collective nuclear sites, and further studies using this method should lead to increased knowledge of them. □

Rising magma

from Peter J. Smith

FEW would now dispute the view that much, if not all, of the volcanic material reaching the Earth's surface originates in the upper mantle. And most Earth scientists would probably agree that magma results largely from the pressure release melting of mantle rock in the rising limbs of convection cells. In the ascending solid state current beneath an oceanic ridge, for example, partial melting of mantle rock occurs, the partial melt fraction becoming the basalt of the new oceanic crust and the residue becoming part of the lithosphere below the Moho. General statements such as these are deceptive, however, implying a degree of knowledge that simply does not exist. The truth is that the mechanism by which magma is transported from the asthenosphere to the Earth's surface is poorly understood, whether the magma rises beneath oceanic ridges, contributes to back-arc volcanism or leads to intraplate eruption.

As far as flow through the lithosphere is concerned, the simplest approach is to assume that a channel exists for the magma; but that begs the question of how the channel first

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formed and fails to relate the continuous flow of magma through a channel to the observed episodic nature of surface volcanism. One of the rather more sophisticated models has a magma body, or 'bubble', floating upwards through the lithosphere (regarded as a highly viscous fluid); but the thermal requirements of this process have not yet been worked out. Whatever the problems may be here, however, flow through the lithosphere can only occur if the magma reaches the base of that layer in the first place; and it is this initial migration of magma in the asthenosphere (where the magma is produced) which is now the subject of a modelling study by Turcotte and Ahern (*J. geophys. Res.* **83**, 767; 1978).

In the Turcotte-Ahern model the asthenosphere is regarded as a porous medium, the mass being convected within it is assumed to rise at constant velocity, and the material of which it consists is supposed to contain a relatively low melting temperature component. As the mantle material convects upwards towards lower pres-

sure regions the lower melting temperature component melts, producing liquid (destined to give basalt at the Earth's surface); the remaining solid deforms to accommodate this liquid, thus producing what is in effect a saturated porous material; and as more liquid is produced it begins to form interconnected channels, thereby providing the material with permeability.

The liquid-produced permeability is central to the model; and Turcotte and Ahern devote much of their mathematical argument to the use of this property in determining the structure of the melting region within which the liquid is produced and in elucidating the relationship between such parameters as mantle ascent velocity; magma viscosity, volume fraction of melt and magma migration velocity. The essential result, however, is that in the permeability model the melt rises much faster than the ascending convective mass. In other words, the permeability arising from partial melting offers a convincing mechanism for magma migration in the asthenosphere. But what happens when that magma reaches the base of the lithosphere is another story. □

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Insect/host plant relationships

from a Correspondent

INTEREST in insect/host plant relations is rapidly increasing with the general acceptance that pesticides are only a partial palliative against insect pests of crops. A recent symposium* on insect/host plant relationships was therefore very timely.

One feature of the meeting was the attempt by physiologists and behaviourists to relate their studies closely to the ecology of insects. This was exemplified in the studies on the Colorado beetle, with T. Hsiao (Utah State University) demonstrating differences in food selection between populations from different regions of the United States. Geographically isolated populations showed considerable differences in both host plant affinity, and in adaptability to different species of Solanaceae. The evidence strongly suggests that specific host-adapted populations are developing quite quickly in North America, and that such biotype differences must be preceded initially by geographic isolation. J. H. Visser and W. C. Ma (Agricultural University,

Wageningen) reported on their detailed study on the odour of potato plants and the manner in which the insect can detect and respond to it. Odour from potato plants elicit anemotactic responses in the Colorado beetles, but none of the individual components applied singly is attractive. A mixture of components with the appropriate relative proportions is required to elicit the behavioural response. An analysis of neurophysiological responses from antennal olfactory cells, showed that discrimination of mixtures of plant volatiles is possible on the basis of information from a limited number of receptor types with different sensitivity spectra.

The attendance of a number of plant chemists reflected the recent expansion of interest in phytochemistry and cooperation between chemists and entomologists. G. Cooper-Driver (University of Boston) in discussing the chemical defences in ferns, emphasised the importance of these plants in the evolution of phytophagous insects and the relative paucity of information on insect/fern relationships. The more ancient orders of insects, however, are

generally better represented than expected, at least on tropical ferns, suggesting a long association between ferns and these orders in spite of the variety of generally deterrent compounds.

R. Hamilton (Liverpool Polytechnic) discussed the basis of attraction of frit flies to oats and showed that there is a synergistic interaction of a lipid and a non lipid material. The lipid was shown to be a hydroxybetadiketone.

An important development was the greatly increased awareness of the value of an evolutionary approach in understanding the present-day relationships of insects with plants. V. Labeyrie (Francois Rabelais University, Tours) emphasised that the insect/plant relationship was not a simple relationship of an insect with its host, but was influenced by other features of the environment. For example the celery fly which mines in the leaves of Umbelliferae also require trees where the sexes meet and mate and must have evolved in a landscape where the two types of plant coexist. E. A. Bernays (Centre for Overseas Pest Research, London) described experiments which demonstrated the capacity of locusts and grasshoppers to feed without ill effects on plants containing high levels of tannins which are general protein precipitants. She considered that the early phytophagous insects had the capacity to tolerate or counter these chemicals, emphasising the dangers of generalisations based on limited numbers of species.

The long term chronic effects of plant chemicals were discussed by a number of participants. Notable in this respect was the contribution by J. M. Scriber (University of Wisconsin, Madison) with an impressive volume of data on a range of lepidopterous species. From the study of over 700 insects from 22 species, he has shown that consumption rates, conversion efficiencies and hence growth rates of tree feeders were lower than for herbaceous plant feeders. This relationship is at least partially a result of the higher water content of herb leaves. □



A hundred years ago

PROF. VIRCHOW has decided to resign his seat in the German parliament. He takes this step solely because his parliamentary duties interfere with his scientific labours; and while he may be a good enough politician, he thinks himself a better savant. From *Nature* **18**, 11 July, 290; 1878.

*Held on 4-9 June, 1978 at the Fulmer Grange Conference Centre, Slough. The proceedings will be published as a supplement to *Entomologia Experimentalis et Applicata*.

review article

Pulsating aurora

A. D. Johnstone*

The aurora is caused by showers of atomic particles in the upper atmosphere, but there are uncertainties about the particles' movements just before they strike the atmosphere. The mechanism responsible for the pulsating aurora would help us to understand what happens in those last few seconds.

THE aurora can be one of nature's most dramatic spectacles. Its size, the rapidity of movement, the sheer dynamism of the display is on a scale beyond any other human experience. It is more cosmic than terrestrial, yet unlike any truly cosmic event it can be appreciated without the aid of any tool other than the human eye. There has been speculation about its causes for centuries. The ideas range from mythical to optical or electrical in character. No-one still believes it is caused by the reflection of sunlight off the polar ice any more than they still believe in a flat Earth. It is now known to be caused by showers of atomic particles, mostly electrons, in the upper atmosphere but there remain many uncertainties about the particles' movement in the few seconds before they strike the atmosphere.

One special type of auroral display is considered here; it is not usually the most spectacular; the intensity is weak and there are no rapid movements, or swirling curtain-like forms. It is intriguing and seems to beg for a scientific explanation when its complex shapes blink on and off as they drift slowly across the sky.

Previous reviews¹⁻⁵ have concentrated on optical observations of the phenomenon. Here we adopt a more comprehensive approach and try to find out the characteristics of the precipitating electron beam and hence deduce the events of those last few seconds.

Pulsating aurora is a natural example of an important class of plasma phenomena—the loss of particles from confinement in a magnetic mirror. The loss process does not seem to be caused by external disturbances but is inherent in the characteristics of the trapped plasma. An understanding of pulsating aurora could therefore have benefits in a wider field than its immediate context, the physics of the Earth's magnetosphere.

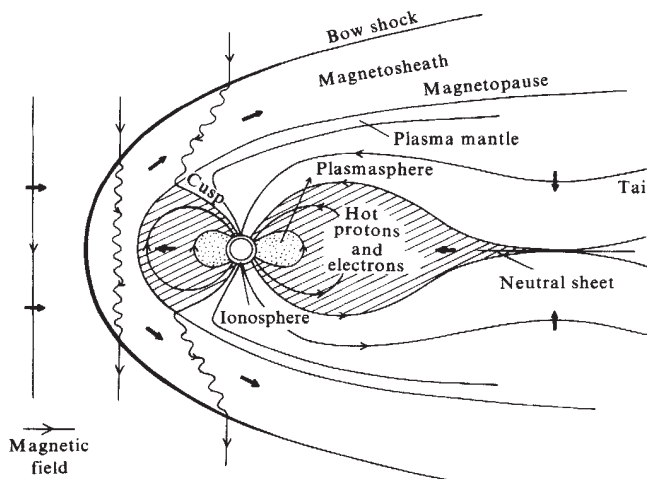
Physical framework of the magnetosphere

All the events described here take place within the Earth's magnetosphere, which is a cavity carved out of the highly electrically conducting solar wind by the Earth's dipolar magnetic field. It is approximately hemispherical on the solar windward side and has a long cylindrical tail extending many millions of kilometres downstream. The diameter varies, but averages 40 Earth radii. Its internal architecture is controlled by the magnetic field which is the sum of the dipolar field, the field of the currents induced in the boundary between the solar wind and the magnetosphere, and various currents flowing in the plasma contained in this volume. The structure is shown in Fig. 1 in a slice through the noon-midnight meridian.

Auroras are seen in a ring around each geomagnetic pole called the auroral oval. The oval is magnetically conjugate with the boundary between the open field lines which stretch into the tail and the region of hot plasma trapped on closed, dipole-like, field lines. The boundary is not fixed in space but moves continually in response to competing internal and external processes which are still not understood. Energetic particles on closed field lines are removed from their trapped orbits by various mechanisms, some of which accelerate them, and precipitated into the atmosphere to create auroral displays. Our aim is to discover the mechanism responsible for pulsating aurora.

Each point in the atmosphere is linked to a field line so that a two-dimensional map of the aurora shows the product of the distribution of plasma in the magnetosphere and the precipitation processes acting on it. Figure 2 was obtained using a satellite travelling northwards over North America and recording the aurora beneath it. It shows the expansion phase of an auroral substorm. The aurora has intensified and became active near local midnight (the right side of the picture) and, at the time of the picture, is spreading polewards and westwards around the auroral oval. Gradually, over the following 30 min

Fig. 1 A schematic view, in the noon-midnight meridian, of the magnetic structure of the magnetosphere. The region containing hot plasma trapped on closed field lines, from which the aurora is produced, is shown cross-hatched. Solid lines are magnetic field lines; short arrows indicate plasma flow.



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or so, the aurora will fade and then perhaps reform as an east-west aligned arc. This substorm sequence imposes a temporal and spatial framework on all auroral events.

The particles' magnetic moment, $\mu = (\frac{1}{2}mV^2 \sin^2(\alpha))/B$, where α is the pitch angle of their spiral motion in the magnetic field, is a constant of their motion along the field line. They are reflected by an increasing magnetic flux density when $\sin^2 \alpha = 1$. At any point S on the field line the pitch angle of a particle which will mirror just above the atmosphere where $B = B_a$ is given by $\alpha(S) = \sin^{-1}(B(S)/B_a)^{1/2}$. Particles with smaller pitch angles will strike the atmosphere before being reflected and thus be responsible for exciting an auroral display. If the intensity within this loss cone around the magnetic field direction is lower than outside it is the sign of a trapped particle population whose lifetime exceeds many bounce periods and hence is also a sign of closed field lines. The bounce period of a 10 keV auroral electron is of the order of 4 s.

As energetic particles precipitate into the atmosphere they ionise and excite atmospheric molecules which then radiate the characteristic auroral emission lines. It has been speculated that other excitation processes, not associated with energetic particles, may occur, but we assume that pulsating auroras are caused by the precipitation of electrons from closed field lines, and that the pulsations are the result of a modulation of the intensity of the electrons in the precipitating beam. The validity of the assumption is considered later.

Observational methods

The precipitating particles can be detected directly from space vehicles, or indirectly by the effects they cause as they strike the atmosphere. Rockets¹²⁻¹⁶ have proved to be the most useful space vehicle for direct observations because satellites cross a pulsating patch in less than a pulsating period. Pulsations have been observed optically by photometric techniques⁹⁻¹¹ for many years but these are now superseded by low-light-level TV cameras⁶⁻⁸ which record both the spatial distribution and the temporal variation. The precipitating particles generate bremsstrahlung X rays which can be detected by balloon-borne instruments at 30 km altitude¹⁷⁻²⁰ and they increase the ionisation in the ionosphere which can be measured by radio methods²¹⁻²⁵.

Small-scale morphology

To be able to use data from different sources, often collected without reference to the visual appearance of the aurora, there has to be some objective selection criterion. Pulsating aurora is a temporal variation in an auroral form; that is, it is a change in brightness, not a change in shape. Therefore, the obvious feature to use, and one which has the additional benefit of being independent of the technique, is the temporal structure of the pulsations^{6-8,10,19,26-39}.

Visible pulsating aurora can be described as a quasiperiodic fluctuation in brightness with periods in the range 2-20 s (Fig. 3). It is quasiperiodic because, in a single train of pulsations, the spacing between successive maxima is not even but varies noticeably from one pulsation to the next. The range 2-20 s has been chosen because there are temporal changes in the aurora with both shorter and longer periods which seem to have different characteristics. Observations with temporal structure falling within the chosen definition apparently form a uniform data set and are concerned with a single magnetospheric process.

The light intensity does not vary in a sinusoidal way, even in a single pulse. Instead it seems to change rapidly from a low state to a high one and back again as if it were switching on and off. The duration of the on and off states varies but the average period is 8 ± 2 s. Another periodic fluctuation, at a frequency of 3 ± 1 Hz, is sometimes superimposed on the main pulsation with a modulation depth usually less than 20% that of the main pulsation^{7,18,26-28}. It is common, but not universal, in pulsating forms seen in the midnight and morning sectors.

Pulsations are weak compared with the quiet arcs and the brilliant breakup aurora. The intensity at 427.8 nm is always less than 10 kR compared with bright forms which reach 100 kR and is usually only just above the visual threshold of the eye of approximately 1 kR. This enhances the switch-on, switch-off appearance to an unaided observer because the light level drops below the visual threshold when it goes off. The amplitude of the pulsation can be several times the background level but has also been detected photometrically with an amplitude of only a few percent²⁶.

The shapes of the forms are extremely varied^{6,27}; they include thin arc segments aligned east-west, featureless patches, patches with intricate detail, and every possible gradation in between. The shape, including intricate detail, persists for many pulsations and only changes on a time scale of minutes. Neighbouring patches pulsate quite independently of each other in phase and period^{6,7,27} may have a dark boundary between them⁷. They merge into each other over tens of seconds. The size is as variable as the shape^{6,7,26,29,30,37}. Coherent pulsations have been detected extending over

Fig. 2 A photograph of an auroral breakup taking place over North America (see text) taken by a DMSP satellite of the US Air Force. The lights in the lower half are the cities of US and Canada. The brightest near the right-hand side is Minneapolis and San Francisco and Los Angeles can be seen in the lower left hand corner.



100 km. There is no statistical survey of the size distribution but it is probable that most patches are less than 50 km across.

All the forms in a display drift across the sky at nearly the same velocity ($100\text{--}1,000\text{ m s}^{-1}$) in a predominantly easterly direction after magnetic midnight^{6,7,27}. There are no strong velocity shears either within patches or between patches, such as are found in more active displays¹. This implies that there are no strong electric field gradients associated with a net charge distribution in the precipitating beam.

The switching on and off, may take place simultaneously over the whole form or it may spread outwards from an initial point within the form. This effect has been called streaming^{27,28}; it is quite a common but not a universal feature of pulsating aurora.

There is also tremendous variety in the shape of a train of pulsations¹¹. The observations are complicated by the tendency of the form to drift out of the field of view. They can start in the low state and end high, vice versa, or any other combination. They may include only one pulsation or many. They can have a steady amplitude or vary in amplitude. There do not seem to be any distinctive relationships between the main parameters, such as size, shape, period, or drift velocity.

Large-scale morphology

If the characteristic period of pulsations was a function of a geometrical property of the magnetosphere, such as the length of a field-line, then one would expect to find a variation in the mean period with geographical location. A dipolar field line at 70° latitude is 50% longer than one at 65° and in the distorted geomagnetic field the relative increase is even longer. Yet there is no strong evidence for a systematic change in the period with latitude, local time or any other variable. Only at Spitzbergen⁴⁹, which is well to the north of the normal auroral zone at 75° , does the mean period seem to be significantly longer. A weak variation may exist but it will require a more extensive survey than has been carried out so far to establish it. The lack of evidence for a systematic variation, together with the facts that the mean periods of adjacent pulsating patches can be quite different and that even in the train from a single patch the period varies, discourages more extensive investigations and leads to the conclusion that the mean period is not related geometrically to the magnetosphere^{6,29,31}. This rules out the possibility that it is connected with the bounce period of the trapped electrons, which is comparable to the periods of pulsations.

The most important large-scale feature in the distribution of pulsations is their relation to the auroral substorm. A casual observer of pulsating auroras soon notices that they can be seen after an auroral breakup in the midnight sector of the auroral oval. It is difficult to put such observations on a sound statistical basis because there is a tremendous range in the size of substorms and they tend to overlap spatially and temporally. The problem has been approached in two ways; by forming statistical distributions with respect to a single parameter such as the level of magnetic activity or local time, or by case studies in distinct auroral substorms. In both types of study the dependent variable is the occurrence of pulsating forms. This parameter may reflect either the actual occurrence probability or the mean amplitude of the pulsations as all the techniques have some amplitude threshold for detection which is usually not clearly defined.

In local time, pulsations are most likely to occur between midnight and 10.00 h (including data from balloon-borne X-ray detectors to cover daylight hours) and the probability of occurrence increases with the level of magnetic activity which is itself an indicator of substorm activity. They have been detected at magnetic latitudes from 60 to 75° usually on the equatorwards side of displays⁴¹. Pulsations have also been observed in the evening sector where they are smaller in amplitude²⁶.

Case studies of the distribution in individual substorms have been carried out and there seems to be fair agreement between them on the principal features^{27,40-43}. The pulsations

appear after an auroral breakup (the expansion phase of the substorm), following in the wake of the polewards expansion of the aurora and also spreading outwards from the region where the breakup initiates towards the Equator and the morning sector. Even the weaker pulsations in the evening sector seem to occur only after the expansion phase. The pulsations persist throughout the recovery phase until a new quiet arc system begins to form.

Relation to geomagnetic micropulsations

It is difficult to establish the exact nature of the relationship between auroral pulsations and fluctuations in the Earth's magnetic field, called micropulsations, because while optical devices isolate individual patches magnetometers integrate the effects of all the pulsations within range. A detailed correlation is impossible except in simple cases^{9,22,32,50-56}.

Auroral pulsations are found to be correlated with micropulsations in the category Pi 1 (irregular pulsations with periods 1-40 s). At the simplest level the two effects have been found to occur at the same time^{32,50}. In more detail, the dynamic frequency spectra of auroral light fluctuations⁹, X-ray fluctuations⁵¹ and ionospheric absorption⁵² fluctuations have each been shown separately to be very similar to the spectrum of micropulsations recorded at the same time. In some cases a detailed correspondence has been found between optical and magnetic fluctuations.

There must be a physical link between auroral and magnetic pulsations but it is not clear whether one is the cause of the other or both are the consequence of some third process.

Measurements of the precipitating particles

The three-dimensional velocity distribution of the precipitating particles^{12,16,45,57-64,67} has axial symmetry about the magnetic field direction and can be presented in two-dimensional form as an energy spectrum, integrated over a range of pitch angles, together with a series of pitch angle distributions at fixed energies. This is an adequate description for pulsating auroras which do not give the dramatic changes in pitch-angle distribution with energy to be found in more active forms. The energy spectrum and pitch-angle distribution can be discussed separately.

The electron spectrum has been measured directly from rockets and can be represented quite well by a Maxwellian form^{12,63,65}

$$J(E) = AE \exp(-E/E_0) \text{ electrons cm}^{-2} \text{ sr}^{-1} \text{ keV}^{-1} \text{ s}^{-1}$$

At high energies ($E > 25\text{ keV}$) this form tends to underestimate the actual fluxes. The measured values of the temperature E_0 lie in the range 2.5-8 keV. During a pulsation the temperature increases by between 0.5 and 2.0 keV but there is very little change in the intensity factor A . The electron distribution seems to be compressed adiabatically during a pulsation and then to expand back to its original state^{63,65}. There is no sign in this spectrum, of acceleration by a parallel electric field such as a strong energy peak accompanied by a field-aligned angular distribution.

The small data set obtained by rockets can be supplemented by several optical techniques which can derive relative changes in the electron spectrum indirectly. They all depend on the variation in height of an auroral form with the energy of the precipitating electrons. The higher the energy of an electron the lower it penetrates in the atmosphere before losing its energy in the excitation of auroral light.

The height of a pulsating form can be measured directly by triangulation from two ground stations. The only feature in the height distribution of the emission which can be reliably identified from both sites is the lower border. The pulsations are found to be lower than other common forms with 80% of a published sample⁶⁶ below 100 km and the lowest penetrating to 83 km. The average height of all auroral forms is above 100 km. The height distribution of light emitted produced by a Maxwellian spectrum has been derived⁶⁸ theoretically. Using

these calculations, and assuming that the lower border would be where the volume emission rate drops to 10% of its peak value, we find that Maxwellian spectra with temperatures of 2 keV and 10 keV produce lower borders at 100 km and 83 km, respectively. The rocket spectra therefore agree well with measurements of the height of the lower border. The triangulation technique does not give the height distribution, which should extend over 30 km at least, nor does it give the height at the pulsation minima where there is no form to be recognised. Other optical techniques depend on averaging over the whole height distribution and can give an estimate of the variation in the spectrum. One method is to measure the ratio of the intensities of two of the auroral emission lines, for example, the atomic oxygen line at 557.7 nm and the ionised molecular nitrogen emission at 427.8 nm. The two substances have different height distributions so that the ratio itself is height dependent. Model calculations have related the ratio to the temperature of a Maxwellian spectrum^{68,69}. The ratio varies during a pulsation in the sense that the spectral temperature increases with increasing light intensity. The results of this optical measurement are in fact very close to the values measured by rockets. In one particular case the temperature

increased from 2.6 to 4.0 keV (ref. 63). These results are supported by other indirect methods^{57,70,71}.

The nature of the spectral changes during a pulsation is therefore well established. The pitch-angle distribution, on the other hand, can only be measured directly from rockets or satellites. Optical techniques are completely insensitive to changes in the pitch-angle distribution.

At high energies ($E > 25$ keV) the electrons' angular distribution is found to contain a loss cone at the minimum of a pulsation^{14,16,58}. The intensity increases monotonically from the field-aligned direction up to 90° pitch angle. This indicates that the process takes place on closed field lines. The distribution becomes isotropic at the pulsation maximum. This implies that the pulsation is caused by an increase in the rate at which electrons are being scattered into the loss cone. At lower energies ($E < 25$ keV) the difference in the angular distribution between the minimum and the maximum of the pulsation is not so distinct. Even at the minimum the distribution is nearly isotropic^{12,58,60} although the auroral intensity is then so low that the accuracy of the distributions is degraded by statistical fluctuations. More accurate measurements of the pitch-angle distribution of low energy electrons in the minima are needed

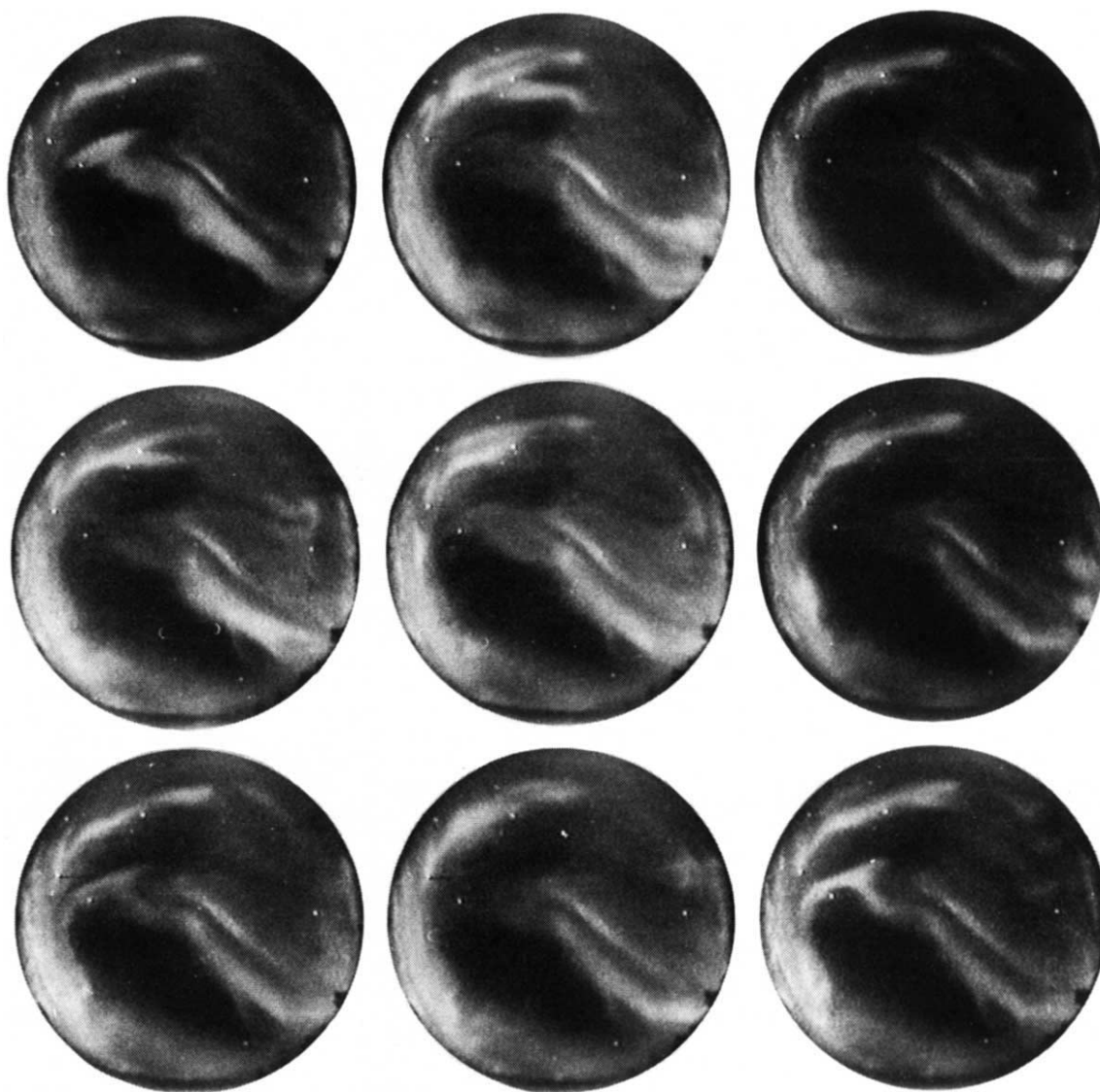


Fig. 3 A sequence of photographs; reading from left to right and then top to bottom, taken at 2-s intervals in a pulsating display. Several independently pulsating patches can be seen. The photographs (from P. Rothwell and R. Thomas of the University of Southampton) are taken from low-light-level television recordings obtained with a fisheye lens directed upwards to view the whole sky.

to establish whether the pulsations can be attributed entirely to an increase in the rate at which electrons diffuse into the loss cone.

If the pulsations were impressed on the beam at some remote point along the field line the fastest electrons in the beam would reach the atmosphere first and cause a phase difference between the pulsations at different energies. Such an effect has been observed several times from rockets^{12,58,59,72} (Fig. 4). Before this can be interpreted as velocity dispersion it must be established that the pulsation observed by the rocket is purely temporal and not caused by movement of the rocket through spatial structure. This has been proved in two ways; by observing the same pulsation from the ground optically^{12,58,60}, and by separating the rocket payload into two packages to make measurements simultaneously at two positions¹². As both packages detected the pulsation at the same time at the same energy it must have been temporal. Then, assuming that the modulation acts simultaneously and at the same place on all the electrons, and that the electrons subsequently follow adiabatic paths in the magnetic field, the distance to the modulation region can be calculated. The validity of these basic assumptions has been confirmed by the internal consistency of the data—when measurements at more than two energies are available, each pair of energies gives the same distance. The results from several rocket flights give distances between 30,000 and 100,000 km from the point of observation. The distance from the rocket to the equator in a dipole field is $\sim 45,000$ km but this should be regarded as a minimum value for the distorted magnetic field in the magnetosphere. Hence, the measurements are consistent with modulation taking place in the equatorial region although not at a simple, fixed location. The distance has been observed to vary systematically from 50,000 to 100,000 km during the course of single flight¹². Other techniques have also detected effects that can be attributed to velocity dispersion⁷³⁻⁷⁵ with source distances ranging from 16,000 km to more than 100,000 km.

Our discussion so far has assumed that all the particles involved are electrons. Certainly they play the dominant role but there are also measurable fluxes of positive ions, mainly protons, to be found and their relationship to the electrons is a vitally important test for the theories. The observation of protons is complicated by the fact that they interact with the atmosphere in a different way from electrons. By the time they have penetrated to 300 km most protons have acquired an electron by charge exchange and are in a neutral state. They are then no longer constrained to follow magnetic field lines. Within a depth of one mean free path for charge exchange they are diffused horizontally by a distance greater than the local atmosphere scale height (> 50 km) (ref. 76). Therefore, below 300 km altitude all spatial structure in the proton beam with scale size ≤ 50 km is smeared out. The hydrogen atoms can be detected by their characteristic radiation in the Balmer β line at 486.7 nm, but as hydrogen atoms they will already have lost the spatial identity of small pulsating patches. It is not surprising that no pulsations have been observed in hydrogen emissions^{35,62}. Only two rockets have been high enough in pulsating aurora to detect protons before charge-exchange interactions. The earlier one only measured proton fluxes at energies above 25 keV, found isotropic pitch angle distributions and no pulsations¹⁶. In a recent flight⁶⁵, proton pulsations were correlated with electron pulsations but delayed by velocity dispersion. The electron-proton dispersion agreed with the dispersion derived from electrons alone, in placing the source near the Equator. The travel time for the protons is approximately 20 s longer than for the electrons which makes it difficult to correlate electron and proton pulsations.

As pulsating auroras occur on closed field lines there should be some activity at both ends of the line of force. It is important to find out whether the pulsations are in or out of phase with each other. It is difficult to identify conjugate pulsations because the size of the pulsating patches is smaller than the uncertainties in locating a pair of conjugate points. There have

been two reports of observations of conjugate pulsations⁷⁷⁻⁷⁹; one was made with airborne TV cameras, the other with ground-based photometers. Both found that the pulsations were in phase. This confirms that the modulation takes place in the equatorial region and also suggests that it works equally well on electrons travelling in the opposite directions along the field line. As the data sample of conjugate pulsations is very small these conclusions must be considered tentative. The modulation at 3 ± 1 Hz, seen frequently in optical emissions, has not been reported in direct measurements of the pulsating electron fluxes. Fluctuations at 3.8 Hz have been found in the intensity of secondary electrons (1–30 eV) in a pulsating display^{80,81}. They are produced in ionising collisions by the precipitating energetic electrons. There were no measurements of primary electrons on the same rocket.

Are auroral pulsations caused by modulated electron fluxes?

So far we have assumed that the answer to this question is 'yes'. The assumption must be critically examined because there are two recent observations which seem to present contrary evidence.

During a series of measurements of the heights of the lower border of pulsating forms it was noticed that the apparent vertical extent of the luminosity was less than 10 km (ref. 82). The heights are consistent with rocket measurements of the electron energy spectrum, as we have already seen, but the measured electron spectrum should give a luminosity profile at least 30 km thick. Even a monoenergetic beam would give a thickness greater than 10 km. It seems therefore that some other excitation mechanism is operating.

Second, pulsations have been either produced or stimulated, within a region of pulsating aurora, by the release of a barium cloud for electric field measurements at a height of 300 km. This suggests that the pulsations are caused by an ionospheric mechanism and not at the Equator⁸³.

There has not been a previous definitive experiment to compare ground-based optical measurements with rocket-borne measurements of the electron spectrum. The principal difficulty is to place the optical instruments at a site where the rocket will cross their magnetic zenith. Then the optical instruments measure the light produced on the same field line as the electrons detected by the rocket uncontaminated by light produced in neighbouring pulsations. As we cannot compare simultaneous observations we must proceed by checking the consistency between different sets of data.

Is there sufficient modulation of the energy flux to cause the observed changes in the optical intensity? Take an observed spectrum¹²,

$$J(E) = 2.2 \times 10^7 E \exp(-E/E_0) \text{ electrons cm}^{-2} \text{ sr}^{-1} \text{ keV}^{-1} \text{ s}^{-1}$$

with E_0 varying from 2.5 to 3.1 keV through the pulsation, assume that the pitch-angle distribution is isotropic throughout and the change in energy flux can be calculated. It varies from 3.5 to 6.6 erg cm⁻² s⁻¹ and would cause a change in the emission at 427.8 nm from 630 to 1,240 R (ref. 69). These are well within the observed range of values. Bryant (personal communication) has reported that when direct comparisons have been possible (never with ideal viewing conditions) the localised electron energy flux fluctuations seem to exceed the line of sight optical fluctuations.

If sufficient energy is available in the electron flux, is it deposited in the atmosphere in the expected manner, that is, through collisions between energetic electrons and atmospheric molecules? The estimate of the electron spectrum obtained by measuring the ratio of two auroral emission lines depends on the average over the entire height luminosity profile and on the theory of auroral excitation by collisional processes. The fact that it produces a result consistent with direct spectral measurements from rockets is strong evidence that the conventional mechanism is operating.

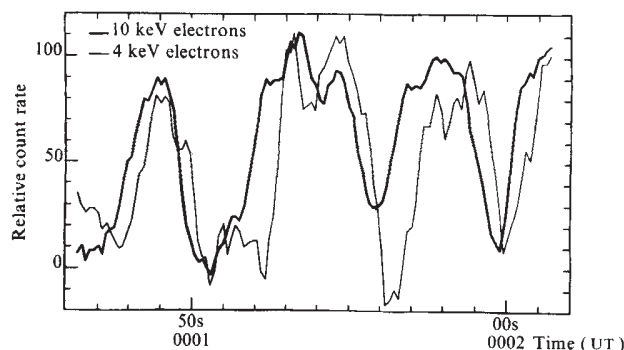


Fig. 4 An example of velocity dispersion in electron fluxes measured from a sounding rocket above a display of pulsating aurora. The variations at 10 keV lead those at 4 keV by approximately 0.55 s which gives a source distance of 55,000 km (ref. 72).

The evidence is strong, but not yet conclusive, that the visible auroral pulsations are caused solely by fluctuations in the precipitating electron flux.

Theoretical mechanisms

The most promising theoretical approach argues that the pulsations are caused by a modulation of the rate of pitch-angle scattering in a diffusion region near the Equator^{84-89,100}. Electrons, in resonance with plasma waves, are scattered without loss of energy into the loss cone. The mean scattering angle is proportional to the wave intensity, assuming many small scattering events in each traverse of the diffusion region. Two extreme situations occur when the mean scattering angle is either much less than, or much greater than the width of the loss cone. If it is greater, than at the exit from the diffusion region, the loss cone will be filled to the level of the intensity outside. Once at this strong diffusion limit the particle flux into the atmosphere cannot be increased and the pitch-angle distribution is isotropic. At the weak diffusion limit the precipitating particle flux is proportional to the wave intensity and the particle intensity increases monotonically inside the loss cone with positive slope. The electron flux can only be modulated by the wave intensity if it is well below the strong diffusion limit at the pulsation minimum.

Two wave modes are known to be important in diffusing auroral electrons. Electromagnetic waves in the whistler mode below the electron gyrofrequency⁹⁰ can resonate with electrons above a minimum energy of the order of $E_R = B^2/8\pi n$ where n is the local plasma density; typically $E_R \sim 10$ keV. The second mode is an electrostatic wave at a frequency above the electron gyrofrequency which⁹¹ resonates with electrons from less than 1 keV up to several tens of keV.

The source of the electrostatic waves is not known although it has been suggested that they are driven by the free energy associated with an observed peak in the perpendicular velocity distribution⁹². The whistler-mode noise can be generated by the anisotropy in the electron distribution created by the existence of a loss cone⁹⁰. The diffusion region for this mechanism is found at the Equator where the greatest proportion of the electron distribution is in resonance with the waves. The wave intensity depends on a balance between the growth rate and the loss through propagation out of the diffusion region. Coroniti and Kennel⁸⁴ showed that in the presence of a micropulsation the growth rate could be modulated by an amount that is exponentially dependent on the amplitude of the micropulsation. They calculated that a magnetic pulsation 2% of the ambient field strength could modulate the precipitating flux by a factor of 2, if the pre-existing diffusion was weak.

This theory makes the auroral pulsations a consequence of magnetic pulsations which, it is suggested, are generated by drift waves^{85,86}. These waves derive their energy from spatial

gradients in the distribution of trapped particles. They have small wavelengths (10–100 km) perpendicular to the magnetic field, and a wave length parallel to the field comparable with the length of a field line. The perpendicular wavelength is comparable with the size of pulsating patches and it has been pointed out⁸⁶ that there could be a relationship between the size of the coherent region of precipitation and the period of the pulsation which would relate the events to drift wave propagation characteristics. There does not seem to have been a serious investigation of this possibility. Possible candidates for the driving source are the temperature gradient at the inner edge of the plasma sheet⁸⁵ and a decrease in the hot plasma density with radial distance from the Earth beyond the plasmapause⁸⁶. Coroniti and Kennel⁸⁵ have calculated that the known temperature gradient could lead to micropulsations with an equatorial amplitude of 1.5%, which is of the right order to produce observed effects. Both the proposed gradients are large scale variations of the plasma parameters and it is not obvious how they could generate a multitude of small asynchronously pulsating patches. The mechanism seems to depend on the characteristics of the plasma contained in the flux tube linked to the individual pulsating patch¹. In the turbulent plasma which is left after the occurrence of an auroral breakup it is possible that intense, small-scale, spatial gradients exist which are able to drive the drift waves.

The theory of Coroniti and Kennel^{84,85} is the only one with any quantitative basis at all. There are necessarily many approximations in their analysis, which may or may not be valid⁸⁷, but they do present a self-consistent and complete picture which may be tested with observations. Although the details may be incorrect, modulation of the rate of pitch-angle diffusion still seems to be the most likely explanation even though there are observations which create difficulties for the theory. For example, the anisotropy of the lower energy electrons at the minimum seems insufficient to allow for the observed increase to the maximum. If protons are able to follow pulsations of the same frequency then it is difficult to account for their variation as pitch-angle diffusion by known waves. The proton gyrofrequency at the Equator is 1.5 Hz which is only slightly greater than the pulsation frequency.

There is one characteristic feature of the morphology of pulsating auroras which seems to have important theoretical implications but which has received little attention. It is the persistence of detail in the shape of a pulsating patch for several pulsations. We have noted that the shape of the patch could reflect either the spatial distribution of the source plasma or the spatial characteristics of the precipitation mechanism. The source plasma, trapped on the field line, is drifting perpendicular to the field at a velocity which is the sum of two principal components. The magnetic gradient drift velocity is proportional to the particle energy and is in a direction which depends on the sign of the charge. The electric field drift velocity is independent of energy and charge. Both components have magnitudes in the range $0.1\text{--}1\text{ km s}^{-1}$ and are directed primarily east-west. The source plasma covers a wide energy range, from a few keV to tens of keV, so that if the source distribution had a spatial shape it would be smeared out by the effects of magnetic gradient drift velocity dispersion within one pulsation or so. As this is not seen to happen then the shape cannot reflect inhomogeneities in the energetic trapped plasma density. Another related argument leads to the same conclusion. If a region of increased plasma density existed, the magnetic gradient drift, in opposite directions for oppositely charged particles would soon separate charges and create an electric field which would then start to distort the shape of the patch itself. However, there are no shear motions in pulsating auroras like those in other displays known to be caused by charge distributions. In a pulsating aurora all the patches drift together at the same velocity. The shape must therefore be related to the precipitation mechanism. Oguti⁷ has suggested that the shapes are those of blobs of cold plasma of ionospheric origin trapped in flux tubes. At their energy ($E < 10$ eV) the

magnetic gradient drift is negligible for both positive ions and electrons. The blobs would drift at the electric field drift velocity without changing shape. The individual pulsating patches would therefore be caused by the presence or absence of cold plasma in the flux tube. Both the wave modes known to cause pitch-angle diffusion of auroral electrons are influenced by the cold plasma density. The resonant energy of the whistler-mode is affected by the total plasma density and the electrostatic mode requires the presence of cold plasma to be unstable. The implications for the pulsating aurora have not been pursued.

The 3-Hz modulation also raises interesting questions because if it were imposed on the electron beam simultaneously with the main modulation at the Equator it would be damped out by velocity dispersion before it reached the atmosphere. It may only affect a narrow velocity range in the beam, which must carry up to 20% of the energy flux or may be the result of an entirely different process. The crucial observations required are measurements of the precipitating electron flux from a rocket during detection of the 3-Hz modulation from the ground.

Related observations in the magnetosphere

The theory of Coroniti and Kennel predicts a relationship between micropulsations, whistler-mode noise and particle fluxes in the equatorial region of the field line. It should be possible to find evidence either for or against the theory from the many satellite passes there have been through this region. The most useful satellites are those in geosynchronous orbits because, according to the best magnetic field models available, they are on field lines which can be traced down to the auroral oval. There are likely to be substantial errors in tracing the field lines because the field models depend on time-averaged data whereas it is almost certain that the actual configuration is time-dependent, particularly during the progress of a substorm.

There have been no reports yet of pulsating electron fluxes within the loss cone (only 1.5° wide) at the Equator although the observation is difficult to achieve. Weak pulsations have been detected in electrons ($E > 50$ keV) well outside the loss cone⁹³.

There have been many surveys of the plasma waves⁹⁴ and both the whistler mode⁹⁵ and the electrostatic mode⁹⁷ have been found to be modulated quasiperiodically in the appropriate range 2–20 s. In one case the modulation of the electrostatic waves was in phase with a magnetic micropulsation.

On two occasions a correlation has been found between micropulsations at the ground and VLF emissions and energetic ($E > 200$ keV) electrons measured from a sounding rocket^{97–99}. The relationship was quantitatively consistent with the theory of Coroniti and Kennel.

Conclusion

When all the evidence is assembled it becomes clear that a great deal is known about the physical process occurring during pulsating aurora. Each of the various instrumental approaches has a significant part to play in building this consistent picture of the phenomenon. There are some important gaps in knowledge which are likely to be filled soon. The Geos 1 satellite now in orbit, and the Geos 2 satellite due to go into orbit next summer carry the ideal instrumentation, and are passing through the most probable location of the interaction region. They will be able to compare electron and proton fluxes in the loss cone, wave intensities in electrostatic and electromagnetic modes over a wide range of frequencies, the variation of the magnetic field strength and the density of cold plasma. If Geos 2 successfully achieves a geosynchronous orbit it will be an opportune moment to attempt correlation with ground-based optical and rocket-borne measurements. When this is done it will also be possible to compare optical and rocket measurements of the pulsations and to make accurate measurements of the pitch angle distribution of the electrons. All these observations are planned to take place during the next year.

The pitch-angle diffusion theory of Coroniti and Kennel has been fairly successful but it is now beginning to look to be in need of renovation. Among the points that need some theoretical attention are: first, the effect of the cold plasma density and its possible influence of the persistence of the shape of pulsations. Second, the existence of similar pulsations in the proton fluxes. Third, whether the apparent temperature variation of the precipitating electrons is physically significant and implies adiabatic compression or is coincidental and caused by the energy dependence of the change in the pitch angle diffusion rate. Finally, the nature of some of the morphological features such as the 3-Hz modulation and streaming.

The stimulation of interest by the new results to be expected in the near future should lead to substantial progress in explaining the phenomenon. It is hoped that this article will help by awakening wider interest and by providing an assessment of the current physical problems.

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articles

Lateral magma flow within rifted Icelandic crust

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Draining of high-level magma reservoirs of central volcanoes during rifting episodes can result in up to 70 km lateral magma flow within the crust and caldera collapse.

ICELANDIC fissure eruptions of evolved magmas are amongst the most voluminous basaltic effusions on Earth. They are characterised by Mg/Mg+Fe²⁺ atomic ratios in the range 0.40–0.47 and have plagioclase, clinopyroxene and olivine as cotectic or near-liquidus phase assemblages. The composition and phase relations of these magmas are incompatible with their direct derivation from the mantle^{1,2} and reflect extensive low-pressure fractionation. In addition, their large volume precludes the possibility that these magmas were stored in dike-like bodies within the crust. A solution to this paradox is offered here by our interpretation that several such fissure eruptions result from the extension of crustal fractures during rifting episodes into high-level magma reservoirs in central volcanoes and the subsequent lateral flow of fractionated magmas into fissure swarms.

The proposed lateral plumbing of magma over horizontal distances of 70 km within fissure swarms has important implications for the context of geochemical sampling of volcanic units in rift zones, as basaltic magmas brought to the surface in some fissure eruptions may accumulate around vents which are geographically widely removed from their source region. Consequently the geochemistry of fissure-erupted basalts cannot necessarily be taken as an indication of conditions of petrogenesis in or geochemical characterisation of immediately underlying mantle.

Three main types of basaltic volcanoes are recognised in Iceland: shield volcanoes, central volcanoes and eruptive fissures. The close association of dyke swarms and central volcanoes observed in the Tertiary of eastern Iceland³ has its

surface manifestation in the relationship between linear fissure swarms and central volcanoes in the active eastern volcanic zone.⁴ The current rifting episode in the northern part of the eastern volcanic zone of Iceland^{5,6} has demonstrated that magma is laterally injected from a high-level crustal reservoir in the Krafla central volcano into a fissure swarm to north and south of the caldera. Björnsson *et al.*^{5,6} have documented a cycle of events in which inflation of a magma reservoir beneath the Krafla caldera takes place over several months. Each inflation cycle ends in rapid deflation, when magma is injected into the fissure swarms up to 10 km north and south of the central volcano. Six such cycles have occurred to date in Krafla since December 1975, and dike formation beneath the fissure swarm is believed to be taking place^{5,6}. In addition a basaltic eruption occurred in the fissure swarm north of the caldera during the tectonic events of 8 September 1977. New insights gained from these events allow a reinterpretation of several older eruptions in Iceland and in particular establish a strong link between high-level magma reservoirs under central volcanoes and some major fissure eruptions in rift zones. We believe that several major fissure eruptions, including the great Lakagígur fissure eruption in 1783 (ref. 7), are a direct consequence of the draining of magma from high-level reservoirs in central volcanoes, as a result of reactivation or propagation of fissures through such centres during crustal rifting episodes. Evidence is presented here for three such proposed connections and the implications are discussed.

The active central volcanoes of Askja, Grímsvötn and Katla are three of the largest central volcanoes in Iceland. Records show that they have frequent minor eruptions of basaltic lava or tephra within or on the periphery of their calderas, with remarkably similar magma composition of successive eruptions in each centre⁸. Askja has a large fissure swarm⁴ and Katla and Grímsvötn also have extensive fissures which occur in the

low-lying ground between the two centres (Fig. 1). Each of these fissure swarms have produced basaltic eruptions with considerable volumes of lava. The Sveinagjá fissure eruption of 1875 (Fig. 2) occurred in the Askja swarm 70 km north of the central volcano. In the eastern volcanic zone three lines of evidence link fissure eruptions to individual central volcanoes. First, the Laki lava flows of 1783 erupted from a fissure striking to the Grímsvötn central volcano and the prehistoric Eldgjá lavas erupted from a fissure trending to Katla central volcano. In neither case, however, can the fissure swarms be directly traced to the centres, as both Katla and Grímsvötn are hidden under extensive glaciers. Second, the major element composition of the basalts from these fissure eruptions is closely comparable to the composition of historic lavas of the proposed source centres (Table 1). For Askja and Grímsvötn and their postulated fissure eruptions we have only compared compositions of lavas which have been generated in a restricted period of each volcano's history. Inclusion of analyses from prehistoric lavas with historic fissure basalts would have little meaning as both these volcanoes have had complex evolutions which are probably associated with long-term variations in magma chemistry. In the case of Katla the temporal control is poor, but the distinctive alkaline character of the pairs of data is evident. We have included all published analyses of historic lavas from the three central volcanoes. We are impressed with the close match in compositions between pairs of fissure-erupted and central volcano basalts. Third, in the case of Askja and Grímsvötn, historical evidence demonstrates contemporaneous activity in the central volcano during the fissure eruptions.

Geological evidence

From early in 1874 to October 1875 a major rifting episode occurred in the northern part of the eastern volcanic zone with both tectonic and volcanic activity occurring in the Askja central volcano and in the fissure swarm to the north of Askja (Fig. 2). The principal volcanic events of this period were a large explosive eruption on 28–29 March 1875 in Askja^{9,10} producing 0.3 km³ (dense rock equivalent) of rhyolite, and a basaltic fissure eruption in Sveinagjá from February to October 1875 in the fissure swarm 50–70 km north of Askja, producing 0.3 km³ of lava¹¹. The eruptions were accompanied by extensive rifting in northern part of the Askja fissure swarm which led to formation of the Sveinagjá graben. The Sveinagjá lava is compositionally very similar to other Askja lavas (Table 1) and to droplets of basaltic magma brought to the surface with rhyolitic tephra in the March 1875 mixed explosive eruption in Askja¹⁰. We have previously proposed¹⁰ that the March explosive eruption was triggered by injection of basaltic magma into a rhyolitic magma body beneath Askja. All of these lines of evidence clearly link the Sveinagjá fissure eruption and the uprise of basaltic magma in the Askja central volcano.

The Sveinagjá–Askja connection is also important for understanding the formation of the 2 km³ Öskjuvatn caldera which formed in the southeastern part of Askja during 1875. There are considerable problems in interpreting the formation of this caldera as being due solely to the emptying of a magma chamber in the rhyolitic equation. First, the 0.3 km³ volume of erupted rhyolitic magma^{9,10} is far too small to account for the 2 km³ volume of caldera collapse. Second, historical evidence¹¹ indicates that embryonic caldera subsidence had started at least 5 weeks before the explosive rhyolitic eruption. Third, the caldera formed in a piece-meal fashion over several months⁹. We propose that this caldera collapse occurred largely as a consequence of the flow of basaltic magma from a high-level reservoir under Askja, into the fissure swarm and the Sveinagjá eruptive fissure. We suggest that the most plausible explanation of the 1874–75 events is that a basaltic and a rhyolitic magma body existed beneath Askja. The onset of crustal rifting resulted in the injection of the basaltic magma into the rhyolitic magma body. Mixing followed¹⁰ triggering the March explosive eruption. Basaltic magma continued to drain from the Askja reservoir until October 1875, feeding the Sveinagjá fissure

eruption, forming an extensive dike, and at the same time causing caldera formation in Askja.

The great Laki eruption of 1783 overshadowed all other contemporaneous events in Iceland, leading to production of 12.3 km³ of basalt lava^{9,11,12}. Published eyewitness accounts show that minor volcanic activity also occurred under the Vatnajökull ice cap, near or within Grímsvötn central volcano in 1783 and 1784 (refs 11, 12). This central volcano has a 35 km² caldera today, rimmed by a 600-m high wall to the south¹³. The caldera now contains a 200 m deep lake which is overlain by a 220-m thick ice cover. All Grímsvötn eruptions are consequently subaqueous phreatomagmatic explosive events such as the events of 1922 and 1934, but not voluminous eruptions.

In May and September 1783 there were reports of an eruption in the glacier in the direction of Grímsvötn. In June of that year frequent earthquakes struck the area and lava started to flow from the 25-km long Laki fissure 45 to 70 km south-west of Grímsvötn at elevation near 600 m above sea level⁷. The initial lava effusion rate of $2.2 \times 10^3 \text{ m}^3 \text{ s}^{-1}$ declined after 50 d as the south-west part of the fissure became inactive, but activity continued on the north-east part of the fissure until February 1784. Rifting during the eruption led to formation of a 300 m wide and 6–8 m deep graben along the central part of the fissure⁷. The final event in this episode was an explosive eruption in Grímsvötn in April 1784 (ref. 10). The compositional similarity of their products (Table 1), synchronous activity and alignment of the Laki fissure (Fig. 1) all point to a close connection with the Grímsvötn central volcano. A hypothetical magma reservoir containing the erupted Laki magma volume (12.3 km³) and having the surface area of the Grímsvötn caldera (35 km²) would have a depth of only 350 m. We suggest that a major collapse occurred in the pre-existing caldera as a consequence of lateral movement of magma in 1783 from a high-level reservoir under Grímsvötn. Today the 600 m high southern caldera wall is a graphic testimonial of extensive collapse in the past.

Fig. 1 Map of southern part of eastern volcanic zone of Iceland illustrating two examples of lateral magma flow. Location of the glacier-covered Grímsvötn central volcano is shown together with associated Laki fissure and 1783 lava. Also shown is glacier-covered Katla central volcano and associated Eldgjá fissure and lava flows. Inset shows locations of Figs 1 and 2 and extent of neovolcanic zones of Iceland.

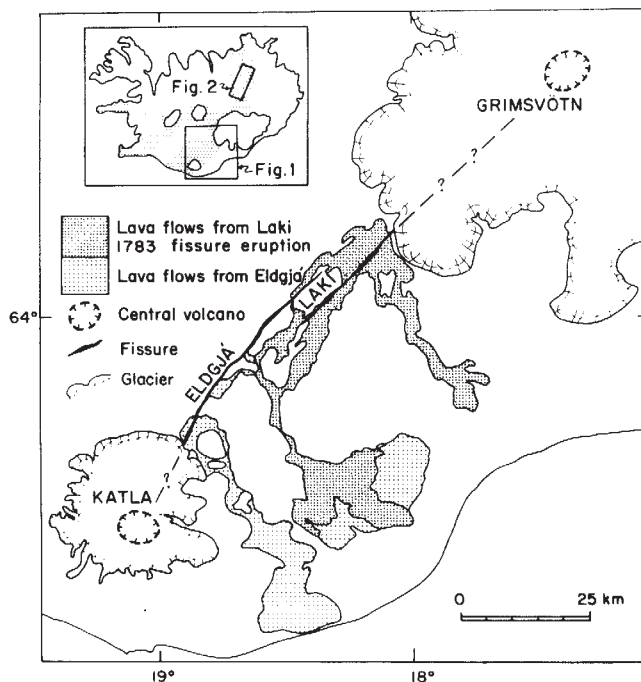


Table 1 Chemistry of basalts from correlated fissure eruptions and central volcanoes in Iceland

	GCV*	LFE†	KCV‡	EFE§	ACV¶	SFE	ACV**
SiO ₂	49.90	49.75	47.35	47.27	50.44	50.04	52.22
TiO ₂	2.74	2.68	4.89	4.27	2.88	2.68	2.24
Al ₂ O ₃	13.50	13.80	13.07	13.98	13.07	13.24	12.98
Fe ₂ O ₃	2.79	2.06	3.50	3.08	2.36	2.36	
FeO	11.88	11.71	12.14	12.20	13.52	13.20	13.96
MnO	0.20	0.22	0.24	0.15	0.26	0.19	0.19
MgO	5.13	5.78	5.32	5.27	5.05	5.46	5.20
CaO	9.73	10.43	9.23	9.92	8.84	9.00	9.29
Na ₂ O	2.84	2.78	2.40	3.08	2.70	3.03	2.57
K ₂ O	0.48	0.41	1.00	0.84	0.64	0.51	0.58
P ₂ O ₅	0.26	0.27	0.23	0.13	0.28	0.27	0.21
H ₂ O	0.64	0.20	1.22	0.20	0.30	0.12	
	100.09	100.09	100.59	100.39	100.34	100.09	99.44

* Grímsvötn central volcano; average of four analyses of tephra from eruptions of 1903, 1922 and 1934 (refs 39–41).

† Laki fissure eruption of 1783; average of four analyses of lava (refs 8, 42, 43).

‡ Katla central volcano; average of two analyses of tephra from eruptions of 1755 and 1918 (ref. 43).

§ Eldgjá fissure eruption; average of eight analyses of pre-historic lavas (refs 21, 43, 45).

¶ Askja central volcano; average of four analyses of lavas (refs 8, 9, 45).

|| Sveinagjá fissure eruption; lava from 1875 (ref. 8).

** Askja central volcano; microprobe analysis of basaltic glass from mixed magma eruption of 1875.

The third case proposed here for a major fissure erupting as a consequence of rifting of a central volcano is the Eldgjá lava in southern Iceland. The 40 km long Eldgjá fissure is in part encased by a graben with maximum depth of 140 m and up to 600 m wide¹⁴. Two major basaltic flows with total volume of 9.3 km³ have issued from the fissure¹¹. One has been dated at 5,000 yr; the other flow may be somewhat older¹⁵. There is, of course, no evidence for synchronous activity in the prehistoric Eldgjá fissure and the Katla central volcano, but the chemical 'fingerprinting' (Table 1) and the trend of the fissure (Fig. 1) are persuasive circumstantial evidence for an intimate connection with the active Katla central volcano, hidden under the ice cap of Mýrdalsjökull. A caldera with a diameter of 10 km has been inferred in the active Katla centre¹⁶. Again lateral movement of such a large volume of basalt in the Eldgjá eruption may have resulted in substantial caldera collapse in the Katla central volcano. The concept of lateral magma flow of the alkalic Eldgjá and tholeiitic Laki magmas readily explains the peculiar juxtaposition of these contrasting basalt types in parallel fissures 5 km apart, and obviates the need for a zig-zag boundary separating the alkalic and tholeiitic provinces of south Iceland¹⁷. Saemundsson¹⁸ has independently noted that lateral magma flow from central volcanoes could account for this feature.

Geochemical evidence

The two principal Icelandic basalt types are evolved tholeiites, mostly quartz-normative, with Mg/Mg + Fe²⁺ atomic ratio of 0.40 to 0.50, associated with fissure swarms and central volcanoes; and second, olivine tholeiites with Mg/Mg + Fe²⁺ in the 0.60–0.70 range and predominantly associated with monogenetic shield volcanoes¹⁹. In the alkalic province of southernmost Iceland¹⁷ these relationships are paralleled by the evolved transitional to mildly alkaline basalts, for example, of Katla central volcano and Eldgjá (Mg/Mg + Fe²⁺ = 0.44) and the Ne-normative alkali olivine basalt shield of Surtsey (Mg/Mg + Fe²⁺ = 0.61).

In the light of the evidence presented above, that fissure-erupted basalts are in many cases derived from central volcanoes, it is now apparent that shield volcanoes and central volcanoes are the loci of major uprise of magma through the Icelandic crust. It is also clear that basalts erupted from central volcanoes and associated fissure swarms have chemical characteristics and phase relations incompatible with their direct derivation from the mantle, as pointed out by O'Hara². Liquids in equilibrium with upper mantle olivine (Mg/Mg + Fe²⁺ = 0.84 to 0.94 (ref. 20)) must have Mg/Mg + Fe²⁺ greater than 0.60,

which is compatible with olivine tholeiites from Icelandic shield volcanoes¹⁹ but suggests extensive fractional crystallisation of evolved tholeiites in central volcanoes (Mg/Mg + Fe²⁺ = 0.40–0.47).

Many of the Icelandic olivine tholeiites are olivine-rich lavas, which have liquidus temperatures in the 1,230–1,250 °C range and have magnesian olivine (Fo_{82–86}) and chromian spinel as liquidus phases¹⁹. Their phase relationships, therefore, indicate some polybaric fractionation during ascent and, together with the chemical evidence, speak for the relatively unevolved nature of these melts. The evolved basalts derived from central volcanoes, on the other hand, have one-atmosphere liquid temperatures in the range 1,120–1,180 °C (refs. 1, 21), with olivine and plagioclase or olivine, plagioclase and clinopyroxene in cotectic equilibrium. Mineral geothermometry with olivine and plagioclase phenocrysts in the Sveinagjá lava gives comparable results, with average olivine crystallisation temperatures of 1,149 °C based on Fe and Mg partitioning²² and average plagioclase temperature of 1,158 °C (refs 23, 24). The basalt magmas erupted in the large central volcanoes such as Grímsvötn, Askja and Katla clearly have compositions and phase relations which testify to extensive modification by low-pressure fractional crystallisation in magma reservoirs. Fractional crystallisation has not, however, proceeded to the point of generation of intermediate and salic magmas in these high-level reservoirs.

Discussion

One implication of our findings concerns the internal structure of central volcanoes and caldera formation. Our hypothesis suggests that magma reservoirs with capacities of the order of 10–15 km³ are an integral feature of evolved Icelandic central volcanoes. Seismic evidence from Krafla indicates such a reservoir at 3–7 km depth²⁵. The configuration of these reservoirs is unknown. They may be cylindrical, lens-like sills, circular in plan and with tapering edges, by analogy with intrusive bodies exposed in deeply eroded parts of the Tertiary of Iceland²⁶. The high magma effusion rates observed from reservoirs, ranging from 200 to 600 m³ s⁻¹ for the Krafla reservoir^{5,6} to 5,000 m³ s for Grímsvötn, as inferred from production rates in the Laki eruption⁷, together with the large total erupted volumes indicate substantial reservoirs. Whatever the shape of the sub-volcanic reservoir, its partial or complete draining is an efficient mechanism for caldera collapse.

Large calderas such as the prehistoric Askja caldera (11 km diameter) do not require large magnitude explosive eruptions of acid magma to form them, but are believed to be a

consequence of drainage of a substantial magma reservoir during a succession of volcano-tectonic events. Even the 1875 Öskjuvatn caldera, formerly regarded as an example of a collapse due to explosive evacuation of a rhyolitic magma chamber, can more easily be explained by drainage of basaltic magma from a central reservoir.

The lateral flow of magma into rift zones to feed fissure eruptions and the accompanying caldera collapse due to magma withdrawal at depth are well-known volcanological concepts which have been largely recognised from research on Hawaii²⁷⁻³⁰. Historical collapses of the Mauna Loa and Kilauea calderas have accompanied large flank eruptions^{27,28}. These collapses have been attributed to flow of magma from reservoirs under the summits into the rift zones²⁸. For example, in the March 1965 eruption of Kilauea $15 \times 10^6 \text{ m}^3 \text{ s}^{-1}$ of lava were erupted from the East Rift Zone along an 8 km long fissure at distances of 15–22 km from the Kilauea caldera²⁹. This eruption was preceded by inflation of the summit and followed by collapse at the summit, demonstrating that the reservoir was beneath the volcano and that the fissure eruption was the consequence of lateral magma flow. The 1968 caldera collapse of Fernandina volcano has also been attributed to magma withdrawal at depth^{32,33}.

Central volcanoes act as the major foci of the ascent of basaltic magma within the Icelandic crust²⁶. There are two principal styles of activity. First, there are small to medium volume eruptions in the central volcano itself, which in the cases of Katla, Grímsvötn and Askja, occur frequently judging from historical records. These eruptions take place on the ring fractures of older calderas or on the fissure swarm within the confines of the central massif. This behaviour may indicate that basaltic magma is being continuously fed into reservoirs within these centres and that the magma periodically takes advantage of pre-existing weaknesses in the volcanic suprastructure.

The second style of activity, already described, is that in which large volume fissure eruptions occur in the fissure swarms adjacent to each central volcano. These events seem to be associated with major crustal rifting episodes. We envisage that during such tectonic episodes compressive stresses are at a minimum, perpendicular to the fissure swarms, and that the magma chamber pressure is sufficient to create fluid-driven cracks which propagate laterally along the fissure swarms from the central reservoir and intersect the surface to produce some Icelandic fissure eruptions and sub-surface dikes. If such reservoirs are at depths of 3–7 km as, for example, in Krafla²⁵, geometrical constraints indicate that the dikes must propagate out laterally but also upwards at a low angle. The two styles of activity are thus related to tectonic events, the 'steady-state' small to medium volume central eruptions occurring in tectonically quiescent periods, whereas large fissure eruptions occur during the major tectonic rifting episodes.

A prominent feature of many Icelandic fissure eruptions, including the three present examples, is the formation of rift zones associated with the eruptive fissures. In many places a distinct graben structure is formed which is generally symmetrical around the eruptive fissures. In Sveinagjá (Fig. 2) historical evidence and geological relationships indicate that the rift zone formed shortly before lava was extruded. Subsidence in the graben reached up to 70 m in the southern part of Sveinagjá. Pollard and Holzhausen³³ have recently developed a theoretical treatment on the mechanical interaction between a fluid-driven fracture and the Earth's surface. Their analysis predicts that in a vertical crack the tensile stresses at the Earth's surface occur at two maxima which are symmetrically located on either side of the surface projection of the crack. The tensile stress is in fact zero directly above the crack. They predict narrow rift valleys above an active dike or eruptive fissure. They further demonstrate a relationship between the depth to the centre of the dike (d), half the height of the dike (a) and the width of the surface graben. For $d/a = 1.25$, the height of the dike is approximately three times the width of the graben³³. Where they are well-developed the Eldgjá, Laki and

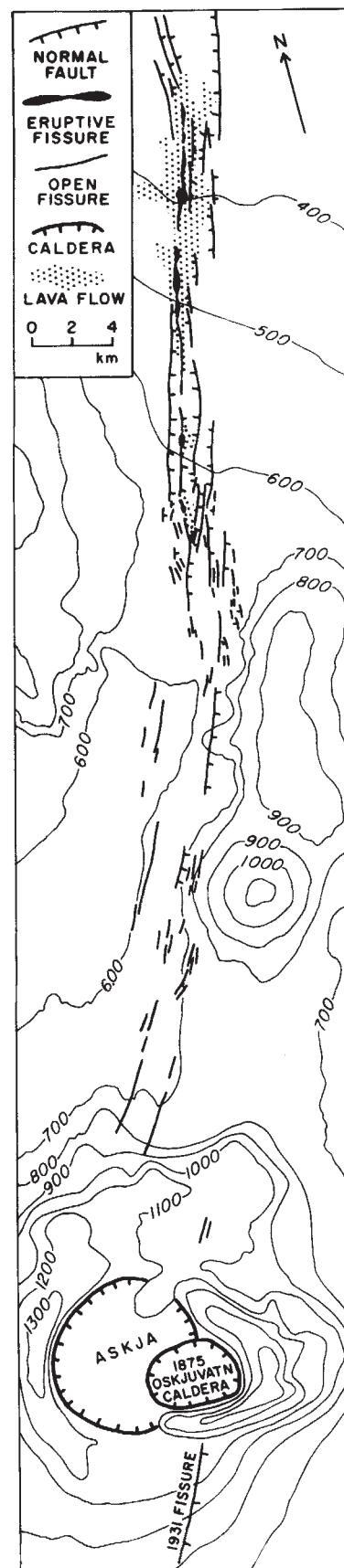


Fig. 2 Map of Askja central volcano, associated fissure swarm and Sveinagjá lava. The prehistoric Askja caldera and the overlapping 1875 Öskjuvatn caldera are located in the south. The Sveinagjá fissure system, which developed during rifting in 1874–75, drained the Askja magma chamber, resulting in subsidence of the Öskjuvatn caldera and extrusion of lava 40–65 km to the north. See Fig. 1 for an index map of the region.

Sveinagjá grabens are 600 m, 300 m and from 1,000 to 1,750 m wide respectively. Pollard and Holzhausen's analysis suggests feeder dikes of 1.8, 1.0 km and from 3.0 to over 5.0 km high for these fissure eruptions. These predictions agree with our view of lateral injection from a high-level reservoir in all three eruptions. In Askja, the wide graben and estimated dike height imply that a much larger proportion of the magma may have been intruded rather than extruded. The total volume missing from Öskjuvatn caldera is 2.0 km^3 of which about 0.6 km^3 can be accounted for by the rhyolitic tephra and the basaltic lava. A volume of 1.4 km^3 can be conveniently accommodated in a dike 70 km long, 5 km high and 4 m wide—dimensions comparable to those deduced from the geological constraints and the theory of Pollard and Holzhausen. In Laki and Eldgjá, a much larger proportion of the magma may have been extruded than intruded.

Finally, we note the regular and approximately equal 40-km spacing of active central volcanoes in the eastern volcanic zone of Iceland (Grímsvötn–Kverkfjöll 40 km, Kverkfjöll–Askja 45 km, Askja–Fremri Námur 44 km, Fremri Námur–Krafla 35 km). A 70-km pattern in spacing of island arc volcanoes has been related to a model of magma generation and diapiric uprise of low-density melt from a buoyant layer at depth³⁴. Mohr and Wood³⁵ also noted a regular spacing of central volcanoes in the Ethiopian rift and drew analogy with Seilig's³⁶ analysis of spacing of salt dome structures. We suggest that the regular spacing of central volcanoes in the eastern volcanic zone of Iceland may similarly reflect the presence of a layer of low-density melt at base of the lithosphere under Iceland. Perturbations leading to sinusoidal deformation of such a gravitationally unstable buoyant layer would be accentuated to form evenly spaced diapiric paths through the overlying lithosphere.

We envisage a steady-state flow of primitive melts (picritic olivine tholeiite or picrite) from base of the lithosphere into channels underlying the active central volcanoes in the eastern zone. These melts will fractionate continuously during slow ascent as well as during their residence in shallow crustal reservoirs to give rise to evolved basalts or quartz-normative tholeiites, erupted periodically and in small volume from the central volcanoes (steady-state style). Independent of the central volcanoes, primitive olivine tholeiite magmas may ascend directly from base of the lithosphere to the surface as a magma-driven hydraulic fracture^{37,38}. Their eruption gives rise

to monogenetic shield volcanoes. Some shield volcanoes, perhaps because of their strategic position within the 40 km spacing pattern, may have evolved into central volcanoes, such as in the case of Fremri Námur.

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Jurassic basin evolution of East Greenland

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The Jurassic basin of East Greenland was formed by rifting from south to north in a series of steps and was filled longitudinally from the north mainly by marine sediments. It may serve as a model for Jurassic graben formation and sedimentation in the areas surrounding the northern North Atlantic Ocean.

ATLANTIC-TYPE continental margins are normally submerged and obscured by thick sequences of post-breakup sediments. This is also the case along the main part of the margins of the northern North Atlantic. East Greenland presents, however, an important exception in exposing a

Mesozoic basin with a length of 800 km, a maximum width of 140 km and containing up to 5 km of Mesozoic sediments. The basin corresponds to the inner parts of the present-day continental margin. Analyses of Mesozoic faulting and sedimentation in East Greenland can, therefore, add significantly to the knowledge of the early stages of this evolution.

Mesozoic faulting

The Mesozoic basin of East Greenland is separated from the western Caledonian highland by a system of roughly N-S orientated dextral *en echelon* faults, which were active during sedimentation and formed, or at least controlled the position of the actual coastlines. There is no evidence for Mesozoic wrenching along these faults, but the *en echelon* pattern may

result from pre-Mesozoic wrenching. The basin is cut by NW–SE trending cross faults situated in and following the major fjords^{1,2} (Fig. 1). The cross faulting which was active throughout the Jurassic culminated in a series of mainly well dated episodes.

Mesozoic faulting is of normal dip-slip type, and a Jurassic–Recent extension of about 6% has been computed³ for the coastal strip in the Wollaston Forland area (block 4, Fig. 1). The southernmost part of block 1 was gently folded in the earliest Cretaceous⁴. The fold axis plunges a few degrees to the south and the uppermost Jurassic and older rocks are folded into three or more low anticlines and synclines over 25 km. The block is about 125 km wide and the folds cannot be interpreted as representing drag effects along a normal dip-slip fault. A possible explanation is that the folding results from compression due to left-lateral wrenching along a cross fault situated in the fjord limiting block¹ to the south.

The Jurassic basin

The Jurassic basin was formed as a N–S trending graben system which opened up from south to north^{1,2}. This took place by down faulting along the N–S *en echelon* master faults and by progressive down faulting from south to north along the cross faults. The individual blocks were thus transgressed in consecutively later Jurassic times in a northwards direction. The down faulted and transgressed blocks on Fig. 1 have been numbered to show the sequence and time when the block was first transgressed. Block 1 is furthest to the south in the exposed part of the graben system (Figs 1, 2), it is the largest block and has the most complete Mesozoic succession. The initial Jurassic transgression here took place in the Pliensbachian (Fig. 2), but the block was a depositional milieu throughout the Triassic and several marine horizons are known from the otherwise fluviatile Rhaetian–Hettangian sequence^{5,6}. To the south on the block the Rhaetian–Hettangian formation comprises alluvial fan sandstones passing into low sinuosity non-braided fluviatile sediments^{5,7,8}. The fluviatile sandstones from the southern area show a strong north–northwest orientated transport direction⁸ roughly parallel with the axis of the depositional basin. To the north, the fluviatile sediments grade into marine sediments. At the northern end of the block tidally influenced deltaic deposits occur, showing an unequally bipolar current pattern with the strongest mode towards the south⁶. This was interpreted as resulting from deposition under tidal activity with a strong southwards mode caused by fluvial overprint. Contemporaneous sediments are apparently absent from block 2 and the existence of a major NW–SE fault zone in the fjord between the two blocks has been suggested elsewhere^{5,9}. Thus the northern part of block 1 was characterised by a southwards palaeoslope and block 2 formed the uplifted northern source area⁶.

The overlying Pliensbachian–Toarcian shallow marine deposits are also restricted to block 1 and Lower Jurassic rocks are absent elsewhere in East Greenland^{1,5}. The sequence commences in foreshore and beach sandstones which pass upwards into tidal, estuarine and shelf mud- and sandstones^{5,10}. Palaeocurrent data are sparse. To the south on block 1, coarsening upwards sequences with N–S to NE–SW bipolar palaeocurrents have been recorded¹⁰. The environment was interpreted as a prograding tidal estuary where shelf deposits pass upwards into fields of migrating dunes and megaripples which accumulated in low relief estuary channel extensions and shoals¹⁰. The interpretation relies on relatively few data but the palaeocurrent pattern conforms with scattered data from the northern part of the block where the dominant transport seems to have been from north to south⁵. Again the sum of data indicates a longitudinal basin fill.

Block 2 was transgressed for the first time between the Toarcian and Bathonian, and a uniform sequence of dark sandy mudstones covered both blocks 1 and 2. The sequence is undoubtedly marine as it contains belemnites and ostrid bivalves but stratigraphically important fossils have not been

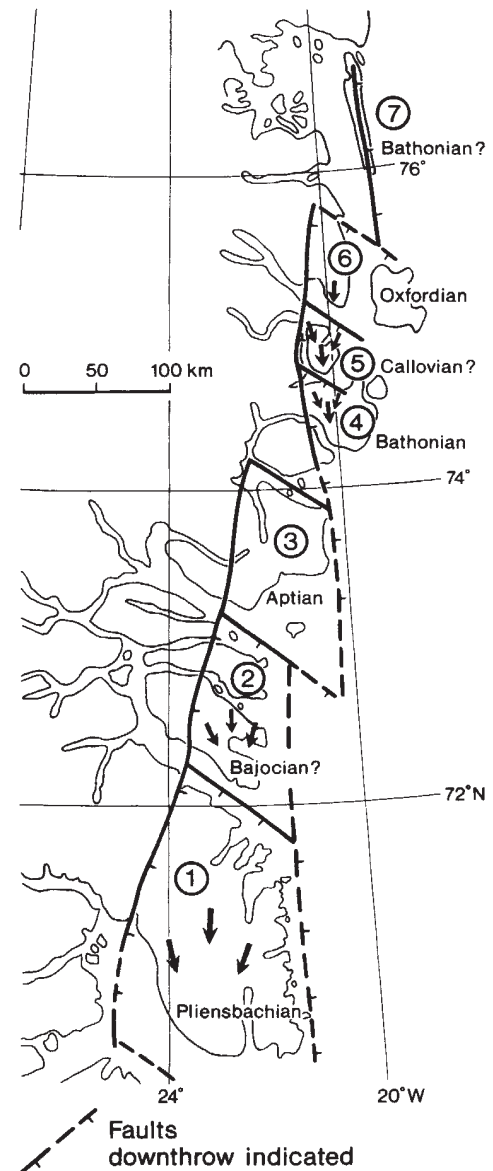
obtained. The age is believed to be Bajocian. The fauna, structures and grain size of the sediments seem to indicate offshore, relatively shallow, restricted marine deposition.

The most important Jurassic transgression in East Greenland took place in the Bathonian or perhaps in the latest Bajocian when fully marine conditions were established on blocks 1 and 2. The northern blocks 4 and 5 were now transgressed for the first time and Middle Jurassic sandstones were here laid down directly on the peneplaned Caledonian basement. Blocks 4 and 5 have yielded few age-diagnostic fossils and it seems as if the transgression is of Middle or Upper Bathonian age on block 4 and of somewhat younger Callovian? age on block 5 (Fig. 2).

Block 6 was first transgressed in the latest Callovian or earliest Oxfordian whilst the northernmost exposed block 7 was transgressed in the late Bathonian or possibly even earlier (Figs 1, 2).

The irregular basement surface of the Milne Land area bordering block 1 to the west was also inundated for the first time in the Bathonian. All previous accounts indicate a Middle

Fig. 1 Map of East Greenland showing the main Jurassic blocks (1–7) described in the text. Arrows show the dominant direction of sediment transport. The time of the first post-Triassic transgression is indicated on each block.



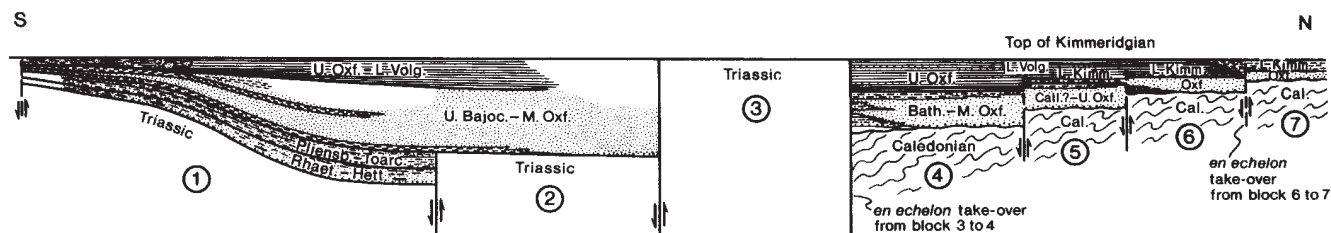


Fig. 2 South to north section through the Jurassic basin of East Greenland. Top of Kimmeridgian used as palinspastic line.

Oxfordian age for this transgression, but new ammonite finds in 1977 have resulted in a total revision of the earlier ideas and the transgression can be safely dated to the Upper Bathonian (T. Birkelund, personal communication).

The Middle Jurassic sediments are uniformly developed throughout East Greenland and are dominated by relatively shallow marine coarsening upwards sequences ending in sandstones or by thick uniform sandy shoreface to beach sequences. The sediments thus mainly reflect deposition by progradation and are of a generally regressive nature. Thin coal seams occur scattered on blocks 4, 5 and especially 6 where they reach a thickness of several metres. Thus, although the Middle Jurassic saw the most important transgression in the Mesozoic history of East Greenland the deposits up to 600 m thick are mainly regressive, recording periods of shoreline progradation.

There are few published palaeocurrent data from the southern blocks whereas the northern blocks 4 and 5 have been studied in some detail¹¹. In block 1, transport is generally to the south⁵ and the sandy member is dominated by coarsening-upwards sequences¹² reflecting southwards prograding coastlines. During progradation the environment changed gradually from muddy offshore shelf to southwards migrating fields of sandwaves and megaripples. Possible exceptions of this longitudinal basin fill are restricted to the western and eastern margins. Bipolar N-S as well as north-west palaeocurrents have thus been recorded from the extreme south-east corner of the block¹³. This might reflect deposition in near-shore and beach environment along the emerged eastern crest of the block. In block 2 the Middle Jurassic sandstones display bimodal N-S and NE-SW patterns with the southern modes being the strongest (L. B. Clemmensen, personal communication). This again conforms with a general longitudinal filling of the basin possibly by southwards prograding coastlines in a tidal environment. The Middle Jurassic of block 4 shows coarsening-upwards sequences to the south whereas the northern sections and all of block 5 display randomly interbedded sandstone facies. Transport is dominantly to the south on block 4, but a tidal influence becomes more pronounced in a northwards direction, and on block 5 bipolar palaeocurrents are recorded with the strongest modes towards south-south-east, south and south-southwest¹¹. No palaeocurrent data are known from blocks 6 and 7 but the facies of block 6 are of a more proximal nature than in block 5 and this tendency culminates in coal-bearing barrier-lagoon sandstones displaying bipolar N-S palaeocurrents¹⁴.

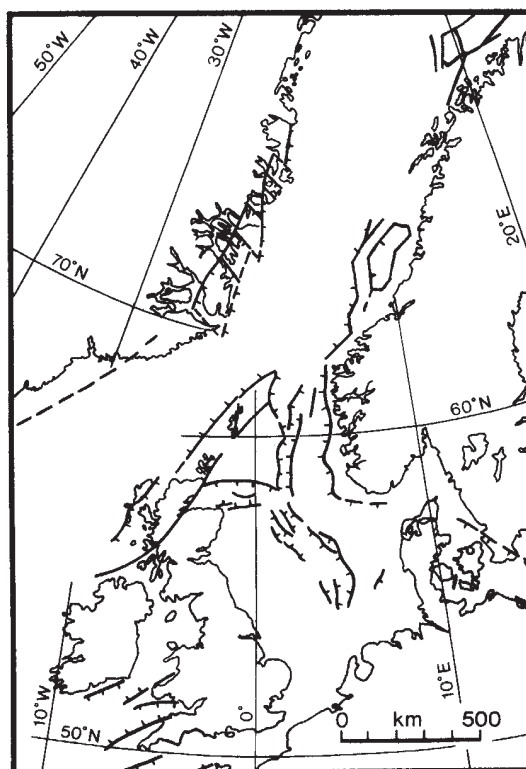
The next major phase of fault-controlled subsidence took place in the late Oxfordian. This phase can be traced all over East Greenland and is shown by an abrupt change from shallow marine sandstones to relatively deep water black mudstones⁵. Only in the northernmost blocks does the sandstone facies persist into the Upper Oxfordian and the shift to mudstones was here delayed until Lower Kimmeridgian^{1,15}. The sedimentation of black mudstones was generally quiet and bears little or no evidence of fault activity. A Lower Kimmeridgian tectonic phase can, however, also be recognised but the evidence for block faulting is in this case mainly stratigraphical¹⁵. An exception is block 1 where well laminated Upper Oxfordian-Kimmeridgian black mudstones contain thick structure-

less channel sands associated with sandstone dykes and sills. The sands were probably emplaced by sediment gravity flows which might have been initiated by earthquakes³.

Directional structures are practically absent from the generally fine-grained Upper Jurassic sediments.

The fault-controlled stepwise submergence of the block-faulted basin culminated in Middle Volgian times with the most important Mesozoic phase of block-faulting in the region. The basin was further fragmented and the depositional regime was profoundly changed. In block 1 and on Milne Land west of this block faulting rejuvenated the source areas and huge volumes of coarse pebbly shallow marine sandstones were deposited during Middle Volgian-Valanginian times. Transport was unidirectional towards south or south-southeast. The facies mainly comprise coarse pebbly shallow-marine sandstones arranged in coarsening-upwards sequences on Milne Land¹⁶ or more randomly interbedded on block 1 (refs 3, 17). There is only meagre evidence for this tectonic phase on block 2, whereas blocks 4, 5, and less important, 6 and 7 contain thick sequences of syntectonic sediments. These northern blocks were fragmented into narrow 10-40 km wide blocks which in contrast to

Fig. 3 The Mesozoic rift of East Greenland shown in relation to contemporaneous rifts in the northern North Sea and North Atlantic Ocean^{21,25-27}.



block 1 underwent strong antithetic rotation towards the continent². The sediments filling the resulting troughs display palaeocurrents which constitute the only case of lateral basin fill during the Jurassic of East Greenland. Sedimentation took place in relatively deep water on coalescent submarine fans formed along the fault scarp and transport is consistently to the east away from the scarps¹⁸. The sediments mainly comprise conglomerates and coarse sandstones which were all emplaced by different types of gravity flows¹⁸. The fault activity of this tectonic phase lasted into Valanginian times, when it finally faded out. It was during this phase that the southernmost part of block 1 was gently folded probably as a result of strike-slip movements along the southern cross fault.

Conclusions

The main Jurassic phases of rifting took place in the Pliensbachian, Bathonian, Upper Oxfordian, Lower Kimmeridgian and culminated at the Jurassic-Cretaceous boundary 135 Myr ago. The directions of the faults were controlled by Caledonian and older structural grain but the rifting was not in itself caused by the older tectonics².

The Cretaceous was tectonically a relatively quiet period dominated by more or less continuous subsidence of the basin and deposition of black mudstones. Initiation of active spreading can be dated to the Palaeocene at the time of anomaly 24 ~55 Myr ago¹⁹⁻²¹.

The East Greenland Triassic-Jurassic rift seems to be part of a three-armed rift system where the two other arms are represented by a little known basin along the coast of Southeast Greenland, and by parts of the northern North Sea graben complex.

If this interpretation is correct, the junction point can be postulated in the area south and south-east of block 1, where the coastline makes a sharp 120° bend². The onshore part of this area is now covered by up to 7 km of Tertiary basalts and very little is known about the Mesozoic geology.

The northern North Sea graben complex never reached the spreading state but seems to have experienced the same structural and sedimentological evolution as the pre-Tertiary East Greenland basin.

The Jurassic basin of East Greenland can thus be compared with the concept of a 'failed arm' of a triple junction²². The rift basin opened up from one end, it was bordered by major faults and it was filled longitudinally by sediment transported down the rift towards the point of where rifting started.

For most ancient rift basins, it is very difficult to demonstrate whether rifting was preceded or accompanied by doming and this is also the case with the Jurassic basin of East Greenland. The drainage pattern, however strongly indicates that rifting was at least accompanied by doming. If doming had not occurred drainage would have been lateral in respect to the rift

and major deltas would have formed at some intervals along the margins of the rift. This is not the case. The drainage system is, on the contrary, longitudinal with the major rivers entering the rift at the points of *en echelon* off-sets of the border faults. This strongly suggests that the main country dip and thus drainage was away from the rift. The situation is probably to some extent analogous to the present-day Red Sea-Nile valley configuration where the doming centred on the Red Sea implies that the major rivers run parallel to the rift. The only place where such a river could flow into the rift would be where the border faults were off-set *en echelon* thus creating a depression in the dome.

It is interesting and perhaps highly significant that the Jurassic cross faults may have influenced the position and direction of the much younger oceanic transform faults and fracture zones. The cross fault between block 2 and 3 can thus be extrapolated into the western Jan Mayen Fracture Zone²³ whilst the cross faults between block 2 and 1 may correspond to a small fracture zone at 70.5°N described by Talwani and Eldholm²¹. Finally, the somewhat hypothetical cross fault limiting block 1 to the south may be extrapolated into the Spar Fracture zone discovered by Johnson and Heezen²⁴.

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Quantum jumps of conductance during formation of membrane channels at cell-cell junction

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The cell-to-cell conductance of a nascent cell junction increases by stable quantal steps. The steps evidently reflect the opening of stable cell-to-cell channels, all alike and growing progressively in number.

CELLS of most organised tissues make junctions that provide a path for direct flow of hydrophilic molecules between cell interiors^{1,2}. The cell-to-cell pathway is well insulated from the exterior³, and, although it has a moderate size limit for permeant molecules, can, nevertheless, exhibit high flow rates.

Thus, the pathway behaves as if made of many parallel aqueous channels¹. A plausible hypothesis is that all the channels in a junction are exactly alike, each channel unit consisting of a pair of permeable elements, one from each membrane, tightly joined in series on the two sides of the junction (junctional unit hypothesis)^{1,4}. It is widely thought that the channels are contained in the intramembranous particles seen in electron micrographs of freeze-fractured gap junctions⁵⁻⁷. The channels certainly do not exist as patent entities in separate cells, but they develop within minutes when the cells make contact. This is clear from experiments in which pairs of cells are manipulated into contact in controlled conditions while their junctional cell-cell permeability is monitored in terms of electrolyte conductance: after a short latency (minutes), a junctional conductance develops, rising progressively from less than $10^{-9} \Omega^{-1}$ to a final steady value of $10^{-6} \Omega^{-1}$ (ref. 8). A simple interpretation of this finding is that the permeable junction develops by a progressive accretion of channels⁸. This interpretation is also consistent with the electron microscopic observation that the number of intramembranous particles in nascent gap junctions increases with time⁹.

On the basis of the junctional unit hypothesis, one would expect the formation of a channel to show itself as a quantal increment in junctional conductance, an increment that might be resolvable experimentally if the channel were sufficiently wide. As probed with fluorescent linear peptide molecules in cells of *Chironomus* salivary gland, the channel diameter is at least 14 Å but not more than a few Ångströms larger (ref. 10 and unpublished data). Such a channel should have a relatively high conductance, perhaps as large as $10^{-10} \Omega^{-1}$ (ref. 4), and so we undertook here to detect quantal changes of conductance during the genesis of a junction. We looked for quantal events during the early phase of junction development, when the resolving power and sensitivity for conductance increments are

greatest²⁹. We induced junction between pairs of cells *in vitro* while continuously monitoring the evolving electrical coupling with a method of high resolution. We found that junctional conductance indeed evolves by a succession of quantal jumps.

The principle of the measurement

Coupling in a cell pair can be represented in terms of the equivalent circuit of Fig. 1. If the nonjunctional membrane conductances, g_1 and g_2 , of the two cells, are of similar orders of magnitude, then, when the voltage displacement in cell 2, V_2 , is several orders smaller than that in cell 1, V_1 , and in the absence of significant capacitive effects, we can approximate the transfer resistance quite well by

$$\frac{V_2}{i} = \frac{g_j}{g_1 g_2} \quad (1)$$

where i is the current passed between cell 1 interior and an extracellular ground and g_j is the junctional membrane conductance. If the measurement is made with sinusoidal current of frequency $(\omega/2\pi)$ high enough to give appreciable capacitive admittance across nonjunctional membranes, the magnitude of the transfer impedance is

$$\frac{V_2}{i} = \frac{g_j}{g_1 g_2 [(1 + \omega^2 \tau_1^2)(1 + \omega^2 \tau_2^2)]^{1/2}} \quad (2)$$

where τ_1 and τ_2 are the nonjunctional membrane time constants of the two cells.

Equation (2) differs from (1) only by a factor that is independent of g_j . In either case, it is clear that, with g_1 and g_2 values stable, V_2 and g_j should increase in direct proportion

Table 1 Changes of transfer resistance during opening and closing of junctional channels

Time (min : s)	Ca ²⁺ injection (pulses)	V ₂ [*] (μV)		n [†]
		Ascending	Descending	
0:00		0.00		0
1:02		0.60		1
1:37		1.25		2
1:58		1.80		3
	5			
2:25			1.35	
2:34			0.60	
2:45			0.00	
3:35	1			
3:41		0.60		1
4:06		1.25		2
	5			
4:31			0.00	
6:30		0.65		1
6:34		1.25		2
7:20		1.80		3
	6			
8:19			1.40	
8:28			0.65	
8:54			0.25	
9:05			0.00	

$V_2^0 = 0.6106 \pm 0.0052 \mu V$. Unitary V_2 increment $\pm$ (s.e. of estimate), evaluated as a least-squares fit of the data of the ascending series by the equation $V_2 = n V_2^0$. $i = 1.00 \times 10^{-8}$ A, r.m.s. (25 Hz sinusoidal). $V_1 = 7.51$ mV.

* V_2 as tabulated here is a constant fraction of true V_2 , which was measured with an undetermined phase error.

† n is the integer that most nearly expresses V_2 as a multiple of a largest common factor.

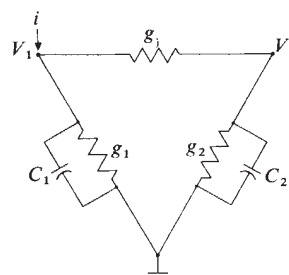


Fig. 1 Equivalent circuit for connected cell pair. Current (i) is passed between cell 1 interior and an extracellular ground, giving rise to intracellular potentials V_1 , V_2 (see Fig. 2). These potentials can be accounted for in terms of junctional membrane conductance (g_j), the nonjunctional membrane conductance (g_1 , g_2), and their corresponding capacitances (C_1 , C_2). The small distributed resistances in cytoplasm and extracellular medium are neglected.

There is no evidence for a significant junctional capacitance.

during junction formation as long as g_j is still small. Hence, V_2 can be used as a direct index of g_j changes, and growth of V_2 by quantal increments would show that g_j also develops by quantal increments. (Even for an equivalent circuit that explicitly includes the possibility of a junctional leak^{8,11} it can be shown that quantal growth of g_j would be the only plausible basis for quantal V_2 .)

The measurement

Cells from *Xenopus laevis* embryos (blastula stages 6.5–8; mostly 8) were isolated in physiological medium (Fig. 2,

legend) containing 2.5 mM colchicine. The colchicine abolished cell movement and division, a condition necessary for stability of cell membrane electrical properties. Permeable junctions formed *in vitro* when we micromanipulated cell pairs into contact. For manipulation as well as for continuous measurements, the cells were kept impaled on microelectrodes⁸ (Fig. 1); cells used had stable membrane potentials ≥ 15 mV (inside negative) and g_1 and g_2 values of similar order in the pair. We chose cells where the values of g_1 and g_2 were stable before cell contact, thus rejecting the great majority of the cells; the g_2

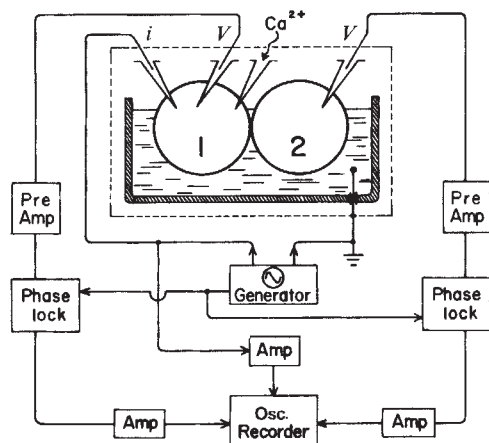


Fig. 2 Diagram of apparatus. The cell pair, in a shielded chamber with inset microscope, is bathed by medium containing (mM): NaCl, 60; KCl, 0.67; NaHCO₃, 2.38; CaCl₂, 0.68; colchicine, 2.5. (Isolation from the embryo in Ca-free, colchicine-free medium.) Cell 1, with diameter ranging 175–300 μ m, contains current-passing (i , 25 Hz, supplied by generator) and voltage-recording (V) 3 M KCl microelectrodes and a micropipette for hydraulic Ca²⁺ injection¹³; cell 2, the smaller of the pair (125–175 μ m), contains a second recording microelectrode (V). Cell 2 is moved into contact with cell 1 by manipulation of the electrode on which it is impaled. The Ca²⁺ micropipette is filled with a solution of CaCl₂ and EGTA, pH 7.0, having free Ca²⁺ concentrations above 5×10^{-5} M, and 0.25 M in fluorescein for estimation of the injected volume²⁶. After cell contact, lock-in amplifiers sample preamplified cell potentials (V) at selectable phase angles with respect to current, and convert them to proportional d.c. potentials. The oscillographic recorder displays amplified potentials and sinusoidal current. Not shown: before cell contact, square-wave generators supply currents to intracellular electrodes for measurement of respective cell input resistances, read on the oscilloscope; lock-in amplifier outputs (after cell contact) are displayed on the oscilloscope as well as on the recorder. The need to move cell 2 towards cell 1 permits only one electrode²⁷ in this cell for g_2 measurement. Holding the isolated cells for impalement was facilitated by letting them settle into shallow pits in a slab of agar at the bottom of the bath (as in ref. 28, but no suction pipette was used).

value was then assumed to remain essentially constant during junction formation. The plausibility of this assumption is suggested by the continuously monitored input resistance of cell 1: cumulative unidirectional changes of V_1 (proportional to $1/g_1$ when $g_j \ll g_1$) never exceeded 0.08% during the events studied.

To follow the course of g_j development, we passed a current (25 Hz; 1.00×10^{-8} A, r.m.s.) between electrodes in cell 1 and the external bathing medium. The major technical problem was to extract from noise a V_2 signal at the microvolt or submicrovolt level. This was solved by driving cell 1 with sinusoidal current and using a phase-sensitive measuring system (Keithley 840) for V_2 . Thus, briefly, from the generator of the 25-Hz sinusoidal current, one derives a voltage of the same frequency and phase, and drives with it a synchronous gating circuit that

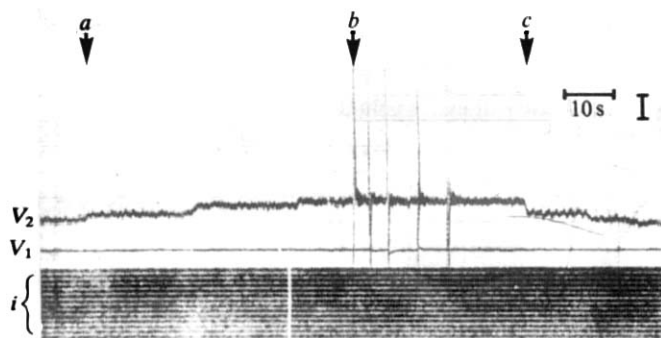


Fig. 3 Oscillograph record of quantal steps in transfer resistance during channel formation and abolition. The high-resolution V_2 record shows a series of three upsteps beginning at a , 1.7 min after cell contact. At b , five successive Ca²⁺ pulses are delivered into cell 1, in the vicinity of the junction, causing three downsteps, which begin at c . (The large spikes in the V_2 trace are capacitive artefacts due to the solenoid that controls hydraulic injection.) Both up- and downsteps are quantal in this experiment (Table 1). The complete data of the experiment to which this record segment belongs are plotted in Fig. 4a. No discrete changes are resolved in the V_1 record at the gain used. Note the recurrent fluctuation of V_2 after the first downstep. Voltage calibration: V_1 100 μ V; V_2 , 3 μ V. $i = 1.00 \times 10^{-8}$ A, r.m.s. sinusoidal.

samples the V_2 signal at a selectable phase ('phase lock'), converting it to a proportional d.c. voltage and excluding incoherent components. The V_2 detector was tuned in phase with the injected current, rather than with V_2 , permitting direct subtraction of a 10- μ V constant error term due to potential drop in the bath electrode. Because of this phase offset, we obtained a proportional measure of V_2 rather than its absolute value. V_1 was monitored by phase lock, but in correct phase. Our resolution for step changes of V_1 and V_2 was 0.8 and 0.05 μ V, respectively; the time constant of the phase-sensitive detectors was 300 ms. The method thus provided an enormous enhancement of V_2 signal over noise, but at the expense of time resolution.

Quantal signals mark the nascent channels

Our basic observation was that during junction formation V_2 rose by quantal steps. Figure 3 shows typical segments of the V_2 (and V_1) oscillograph records after a pair of cells was brought into contact. Shortly after contact, a step sequence appeared on the V_2 record. Figure 4a plots all the V_2 steps recorded in this experiment; the steps were in units of 0.6 μ V or twice that.

Our experimental conditions favoured a relatively slow development of junctional conductance, particularly at onset; here, despite our limited time resolution, the quantal feature was distinctly visible. In the successful cases, a V_2 signal appeared within 0.7–1.5 min of cell contact. In some experiments (not set up for resolving unit increments), the observations were extended for longer periods. Junctional conductance then rose over 0.5–2 h to $10^{-7} \Omega^{-1}$, and more rapidly over the following 1–3 min, to a final maximum of the order of $10^{-6} \Omega^{-1}$.

The discrete V_2 steps were seen during the first 10 min after contact. Here, a criterion was needed for distinguishing junctional channel events from other disturbances of the V_2 record. Given the time constants of the detection system, only relatively persistent shifts in the V_2 records were significant; indeed, discrete, stable V_2 steps stood out clearly from other disturbances. In scanning an aggregate of 10 min of post-contact records of successful runs, we found 14 steps, all integer multiples of the unitary V_2 step of the particular run.

On the other hand, in 40 min of pre-contact records (10 experiments, including all of the successful runs), there were no step events with $|\Delta V_2| > 0.05 \mu\text{V}$ and duration > 2.5 s. Thus, any V_2 step exceeding these limits (after contact) was deemed a junctional event.

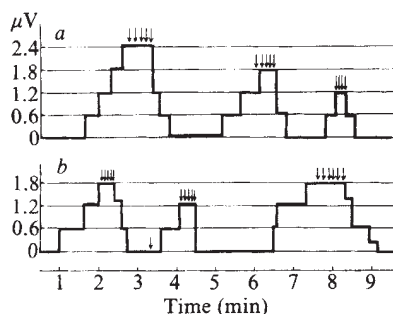


Fig. 4 Time course of transfer resistance during formation and closing of junctional channels. Cell contact is made at 0 min; thereafter, the cells remain in contact, and, except for Ca^{2+} injections (each arrow denotes one injection pulse), there is no further experimental intervention. Upsteps of V_2 occur spontaneously; downsteps, after Ca^{2+} injection only. In *a*, all changes are integer multiples of a unitary value; in *b*, the downsteps are not, in general, integer multiples of the upstep quantum (Table 1).

Table 1 presents the results of one experiment (Fig. 4*b*). All upsteps of V_2 are, to a close approximation, equal. The apparent unitary V_2 step is $V_2^0 = 0.6106 \pm 0.0052 \mu\text{V}$ (by least-squares fit). In four such experiments, there were 24 upsteps; 23 were unitary and one was a two-unit step. The standard errors of V_2^0 ranged from 0.6 to 1.7%. The small standard error in each case shows clearly that the V_2 change is quantal and, hence, that the junctional conductance itself changes quantally.

Ca^{2+} closes the channel

With the recording of such low-level signals, there is naturally a risk of artefacts. Here, we used the fact that junction permeability is sensitive to the cytoplasmic free Ca^{2+} concentration ($[\text{Ca}^{2+}]_i$). The molecular size limit for junctional passage decreases gradually with rising $[\text{Ca}^{2+}]_i$ in the range 10^{-7} to $\sim 5 \times 10^{-5}$ M (ref. 12); at higher $[\text{Ca}^{2+}]_i$, passage is blocked for all molecular species, including the small electrolytes, and so junctional conductance then falls by several orders of magnitude^{11,13}. In terms of the junctional unit hypothesis, this means that the channel closure is graded, becoming complete at the high $[\text{Ca}^{2+}]_i$. Thus, if the observed V_2 steps truly reflect junctional conductance changes, one should be able, by Ca^{2+} injection, to reverse the steps partially or to abolish them totally.

The Ca injections were made deep into the cell and close to the junctional region, avoiding (in successful runs) the effects of Ca^{2+} on nonjunctional membrane. (With injections close to the outer surface, nonjunctional membrane conductance fell.) By such injections, the upstep trend was reversed. The effects of the Ca injections fell into two classes. In one class, V_2 decreased by the same unit steps as those of the rising phase, or by double these amounts (Fig. 4*a*). In the other class, not all the V_2 decrements were integer multiples of the upstep quantum; some, in fact, were smaller (Fig. 4*b*, Table 1). It is possible that these smaller downsteps have a quantal structure of their own, but the data are insufficient to decide this. In either class, the Ca effect was spontaneously reversible; the upstep trend resumed within 1.2–2 min of the Ca injection. The cycle of V_2 abolition and recovery was repeatable (Fig. 4). The local

$[\text{Ca}^{2+}]_i$ elevations in the first class presumably reached the level needed for closing a channel completely, whereas those in the second class sufficed only for closing it partially.

In sum, formation of a permeable junction proceeds as though by accretion of unitary channels; each quantal upstep of conductance represents the complete opening of a cell-cell channel and each downstep a complete or a partial closure.

The present results provide the most direct demonstration of the presence of specialised cell-to-cell channels and the first evidence of their unitary nature. They do not give the absolute value of the unit channel conductance, for, as already indicated, only a proportional measure of V_2 was determined, and g_2 , τ_1 and τ_2 were not evaluated absolutely.

The channels are stable

The cell-cell channels we detect seem to be quite stable in their open state. Once formed, they persist in a conductive state for at least several minutes; closures were not seen unless Ca^{2+} was injected. This contrasts with other membrane channels, for example, the 'K channel'^{14–16}, the 'Na channel'¹⁵, the 'acetylcholine channel'^{15–18}, the 'alamethicin channel'^{19,20} and the 'gramicidin channel'^{21–24}, all shortlived. The stability of the open cell-cell channel implies a stable molecular interaction. This may be a consequence of the paired structure of the channel on the two sides of the membrane junction, a pairing of subunits. Patency stabilisation and structural stabilisation of the channel thus result at once as interactions of the same set of subunits. (This does not preclude separability of the two stabilisations; there are indications that channel closure by Ca^{2+} (refs 11, 13) may not entail gross dissociation of unit channel structure²⁵.)

We do not rule out the additional occurrence of shortlived channel openings. Had conductance states with lifetimes < 0.5 –1 s occurred, they would have been missed because of the time constant of the detector. We did, in fact, detect occasional V_2 transients with half-times of about 1.3 s occurring in short bursts (as in Fig. 3). Four such bursts were found in a total of 10 min of (post-contact) record. These transients may reflect unstable channel openings or closings.

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letters to nature

Is HD26676 an unusual radio star?

STROM AND HARRIS¹ have detected a faint radio source (4 mJy at 6 cm) whose position is coincident, to within an error region of 3×18 arc s, with the 6 mag late B star HD26676 (=HR1307). Although a prominent reflection nebula surrounds this star², they argue convincingly¹ that the nonthermal nature of the radio spectrum suggests identification with the star itself. As Strom and Harris stress, this association would be remarkable, as all previously identified radio stars are radio and optical variables and/or binaries. We report here that no such evidence exists for HD26676 or the radio source.

HD26676 has been classified as B8V by Cowley³ and B7V by Racine². The main sequence assignment by Racine is based on the equivalent widths of Balmer lines. *U*, *B*, *V* colours by Crawford⁴ and Cousins⁵ more closely correspond to B7V with a colour excess E_{B-V} of about 0.15 mag. The distance to HD26676 is ~ 125 pc as given by Racine² who assigns the star to the association Taurus R2. Among known radio stars the eclipsing binary β Per has the spectral type, B8V, most similar to that of HD26676. The orbital period of β Per is 2.9 d, and its distance about 30 pc. Its radioflux is highly variable, from less than a few mJy to 1 Jy (refs 6, 7).

We do not know of any detailed, high dispersion search for radial velocity variations in HD26676. Such observations are reported here which demonstrate that this star is almost certainly not a close binary. The data are based on plates obtained with the Coude spectrograph of the 3 m Shane reflector of the Lick Observatory on five nights in 1977–78. One spectrogram, covering the wavelength range $\lambda\lambda$ 3,500–4,800 with dispersion $16 \text{ \AA mm}^{-1}$, was obtained on each of these nights. Our estimate of the spectral type from these plates

does not agree particularly well with our result, either as to constancy or mean velocity. However, our data are of substantially higher dispersion, on well-widened spectrograms (0.9 mm), and we do not regard this discrepancy as significant.

From the results of Table 1, we tentatively exclude systematic radial velocity variation of amplitude greater than a few km s^{-1} on timescales of 1 day to several weeks, periods relevant for other known binary radio stars. Even a severely undermassive companion would be expected to cause variations exceeding this limit unless the system is accidentally seen nearly pole-on. The rotational broadening, estimated very roughly as 250 km s^{-1} from the weaker lines on our spectrograms, argues strongly against this possibility. This line width corresponds to a rotational period of about 0.6 d. In most close binaries, including β Per, rotational and orbital periods are about equal. We conclude that HD26676 is unlikely to be a close binary.

This constraint makes the result of Strom and Harris¹ more intriguing. Either the positional association of HD26676 and the radio source is accidental, despite the probability of order 10^{-6} quoted for such a coincidence¹, or this object may represent a new class of radio star. A radio error box with an order of magnitude less area will help greatly to resolve this question. Instrumentation currently available is capable of such an observation.

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Table 1 Radial velocities of HD26676

Heliocentric julian date	Radial velocity (km s^{-1})
2443470.824	$+20 \pm 2$ (m.e.)
529.699	$+26 \pm 4$
558.659	$+20 \pm 2$
559.602	$+21 \pm 5$
560.601	$+21 \pm 2$
Weighted mean	$+22 \pm 1$

is B8V. The confluence of the Balmer lines at about $n = 16$ confirms that the surface gravity is that of a main sequence star.

The spectrograms were measured with a Grant-type engine and velocities derived from eight Balmer lines between H γ and H14 on each plate. Shallow lines of HeI and MgII provide velocities consistent with but more uncertain than the Balmer series, and these were not used in the final analysis. A list of the observations and resulting mean velocity for each plate are given in Table 1. These data indicate that the velocity is constant to within the uncertainties. A weak interstellar K line gives a velocity of $+20 \pm 3 \text{ km s}^{-1}$. Four radial velocity measures of this star from two series of spectrograms have also been reported by Hube⁸. Their weighted mean, $+5 \pm 8 \text{ km s}^{-1}$,

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Relative abundance of antiprotons and antihelium in the primary cosmic radiation

THE importance of determining the abundance of antiparticles in the primary cosmic rays has already been discussed¹: for example, Steigman has considered the value of $\bar{p}$ as a probe in distinguishing various propagation models. We report here on the measurement of $\bar{p}/p$ and He/He in the 4–100 GeV/c range made from a balloon flight in May 1976 of a superconducting magnet spectrometer from Palestine, Texas, under 5.8 g cm^{-2} of residual atmosphere. The upper limits derived are the most stringent to date.

We have previously reported on the $\bar{p}/p$ in the 4.2–12.5 GeV/c range from an earlier (September 1975) balloon

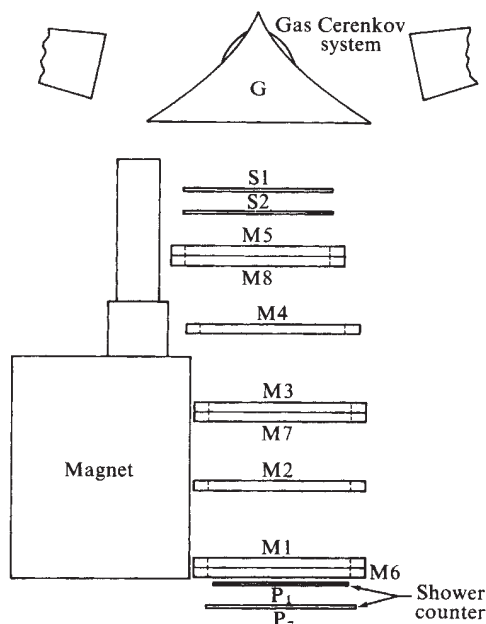


Fig. 1 The magnet spectrometer, S_1, S_2 are charge determining scintillator, P_1-P_7 form the 7.2 radiation length shower counter, G is the air gas Cerenkov counter and M_1-M_8 are position measuring detectors. The line integral $\int B \times dl$ varies from 1 to 9 kGm^{-1} for acceptable trajectories, where $|B|$ is the magnetic field strength and $|dl|$ is the element of path length.

flight³. The present instrument, shown in Fig. 1, differs from the previous experiment in one important respect. In the present flight there were three additional position measuring multiwire proportional counters (MWPCs). The additional MWPCs (M_6, M_7 and M_8) result in greater momentum resolution as well as greater capability for rejecting background events; in all other respects the two flights were identical. The method of data analysis is identical to that reported earlier³.

Figure 2a shows the deflection ((rigidity)⁻¹) distribution of charge $Z = 1$ downwards moving particles. These events were selected to have a single trajectory through the eight MWPC stack, to have no cascade in the 7.2 radiation length lead scintillator sandwich and not to have triggered the gas Cerenkov counter (cut-off Lorentz factor of 40). The plot, therefore, contains background atmospheric (or instrument produced) μ^- , π^- , $\bar{p}$, and very high energy protons that spill over to negative deflection due to the finite momentum resolution of the instrument as well as galactic antiprotons.

The solid line in Fig. 2a is the atmospheric ($\mu^- + \pi^-$) at float altitude and has been obtained by folding in the calculated atmospheric μ^- and π^- deflection spectrum⁴ with the experimental resolution function and the experimentally measured gas Cerenkov efficiency³. The resulting curve has been normalised in the deflection interval -0.9 to $-0.3 \text{ (GeV/c)}^{-1}$, that is, the region in which only atmospheric secondaries μ^- and π^- are expected to be present. The dashed line is the result of combining the experimental resolution function with a proton rigidity spectrum $\propto R^{-2.75}$. The combined ($\mu^- + \pi^-$) background and spill over curve clearly give a good description of the observed data. In particular, above the geomagnetic cut-off of 4.5 GeV/c (deflection $\leq 0.22 \text{ (GeV/c)}^{-1}$) there is no enhancement above the calculated background. We can, therefore, place an upper limit on the antiproton flux. In the deflection interval -0.22 to $-0.88 \text{ (GeV/c)}^{-1}$ (the upper limit is chosen so that spill over proton contribution is negligible) we expect to see 506 events. The excess of $\bar{p}$ is thus $-6 \pm (25)^2 + (22)^2)^{1/2}$ where 22 is the statistical uncertainty on the

observed 500 events and 25 the uncertainty in the normalisation procedure. In the corresponding positive deflection interval ($0.08-0.22 \text{ (GeV/c)}^{-1}$), we observe 95,004 protons. We therefore have $\bar{p}/p = (-0.63 \pm 3.53) \times 10^{-4}$ in the $(4.5-12.5 \text{ GeV/c})$ interval, or a 95% confidence level upper limit of 6.4×10^{-4} . This is completely consistent with our³ earlier results of 6.6×10^{-4} in the -0.24 to $0.08 \text{ (GeV/c)}^{-1}$ interval. The combined upper limit from the two flights is then 5×10^{-4} . These limits are at the top of the detector. As secondary atmospheric $\bar{p}$ production has been neglected they can also be regarded as upper limits at the top of the atmosphere.

The analysis of the antihelium is similar to that of antiprotons except that there are no expected secondary production processes. We examine the deflection distribution of charge $Z = 2$ downwards moving particles, and find no negative deflection events above those expected from the spill over of

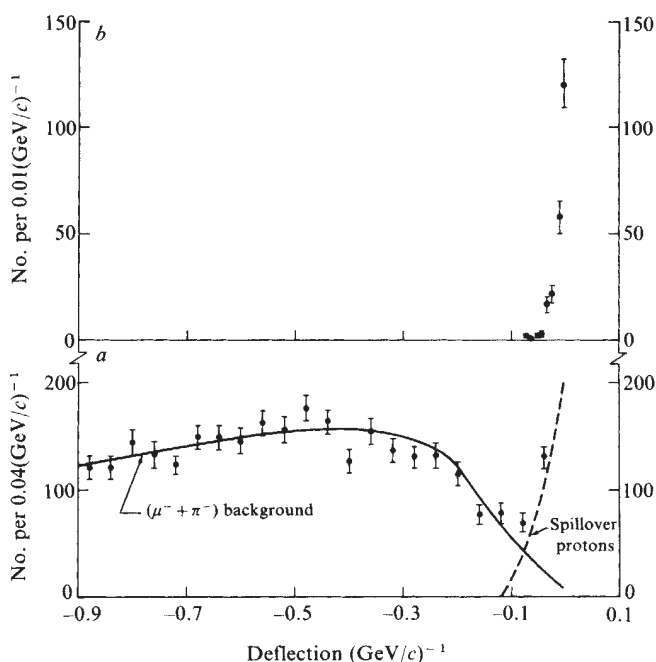


Fig. 2 a, Deflection (negative) distribution of $Z = 1$ (G off), $\int B \times dl \geq 3 \text{ kGm}^{-1}$, downwards moving particles. The solid curve is the ($\mu^- + \pi^-$) background at float altitude and the dashed curve is the spill over high energy protons. b, deflection (negative) distribution of $Z = 2$ (G off) downwards moving particles, all $\int B \times dl$.

high energy helium nuclei. Figure 2b gives the deflection distribution of $Z = 2$ particles which have negative deflection. We see no events in the -0.2 to $-0.10 \text{ (GeV/c)}^{-1}$ interval whereas there are 11,376 helium nuclei for the corresponding positive deflection interval. We therefore have an upper limit of $\bar{\text{He}}/\text{He} < 8.8 \times 10^{-5}$ in the $5-10 \text{ (GeV/c)}^{-1}$ interval at a 95% confidence level. By combining the data from the September 1975 flight, which gave an upper limit of 1.7×10^{-4} , this limit can be lowered to $\bar{\text{He}}/\text{He} < 5.8 \times 10^{-5}$ in the $5-10 \text{ GeV/c}$ interval. Similarly, by examining the data with higher momentum resolution ($\int B \times dl \geq 3 \text{ kGm}^{-1}$), we find $\bar{\text{He}}/\text{He} < 1.6 \times 10^{-4}$ in the $10-33 \text{ GeV/c}$ interval at 95% confidence level. In the $4-33 \text{ GeV/c}$ region we find $\bar{\text{He}}/\text{He} < 1.0 \times 10^{-4}$. This limit is about a factor of 5 lower than that given by Smoot *et al.*⁵ for the same rigidity interval.

The upper limits in the interstellar space would be about 10% higher due to corrections for the material of our gondola and the atmosphere. These data require no correction for any

positive particle spill over in the 4–33 GeV/c interval. By applying a spill-over correction, which is determined by fitting the entire $Z=2$ deflection (-0.2 to $+0.08$ (GeV/c) $^{-1}$) distribution to a power law in deflection, we can extend the above momentum range and obtain a limit of 10^{-2} in the 33–100 GeV/c range.

Our upper limit of $\bar{p}/p < 5 \times 10^{-4}$ in the 4.2–12.5 GeV/c range; $\text{He}/\text{He} < 5.8 \times 10^{-5}$ in 4–10 GeV/c; $< 1 \times 10^{-4}$ in 4–33 GeV/c; and $< 10^{-2}$ in 33–100 GeV/c are the most stringent upper limits so far obtained.

The present upper limit on $\bar{p}/p$ of 5×10^{-4} is only slightly higher than the expected value of 4×10^{-4} for a leaky box-model and nearly equal to the 4.8×10^{-4} expected in the Peter-Westergard model². It completely rules out the closed galaxy model².

The upper limit of $\bar{\text{He}}/\text{He}$ of 1×10^{-4} in the 4–33 GeV/c range is the most stringent ever obtained. Following an argument of Steigman¹ and assuming that the leakage life of the cosmic rays from the galaxy is ≥ 10 Myr, this upper limit on $\bar{\text{He}}/\text{He}$ suggests that the local group of galaxies and possibly the Virgo supercluster contains predominantly normal matter.

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Predicted intensity of the solar maximum

It is desirable to know beforehand, for planning geophysical and other studies, the intensity of a future period of solar activity. Ohl has shown that the level of geomagnetic activity (as indicated by K_p index) in the descending branch of a solar cycle was well correlated with the height of the following cycle and predicted a maximum annual average Wolf's number of 140–180 for cycle 21. Sargent² has shown that a similar analysis using the 'aa' index for the past 100 yr (ref. 3) and sunspot number at the minimum of the cycle gave a multiple regression equation:

$$R_{\max(n+1)} = 3.91 + 8.56 (X_1 - 0.92 X_2)$$

where X_1 is the average value of the monthly mean aa indices for the 36 months preceding $R_{\min(n)}$ and X_2 is the value of $R_{\min(n)}$. Sargent predicted a Wolf's number of 150 or more for cycle 21. We describe here a simpler version of the same method—correlating $R_{\max}$ with the minimum annual aa index in the preceding years.

Figure 1 shows the maximum annual sunspot numbers for the various solar cycles of the past 100 yr plotted against the minimum values of the annual average aa index from the 6 yr immediately preceding each solar maximum. For example, the annual sunspot number in the solar cycle no. 19 was 190.2 in 1957. The annual mean aa indices for the previous 6 yr, 1951–56, were 28.8, 27.9, 22.2, 17.3, 17.6 and 24.8. Among these, the minimum value 17.3 (for 1954) was plotted against the maximum sunspot number 190.2 for 1957 (actual values are

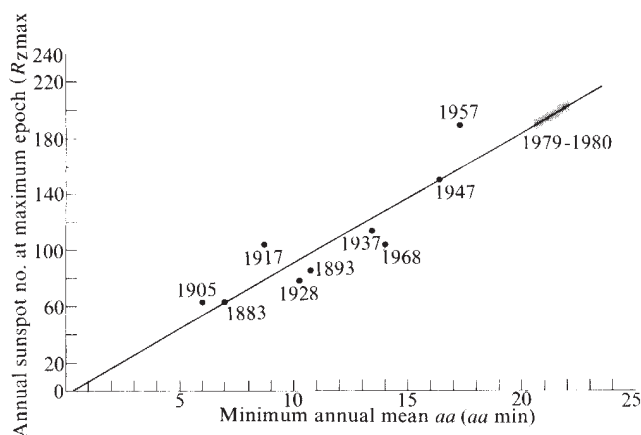


Fig. 1 Annual sunspot no. at maximum epoch plotted against preceding annual minimum aa indices.

given in Table 1). The correlation coefficient is reasonably high (+0.89) and the regression line obtained by the least square fit is:

$$(\text{Sunspot no.}) = (9.4 \pm 1.8)(\text{aa index}) - (2.9 \pm 21.3)$$

To apply this relationship to cycle 21 which is just beginning, we note that the annual aa index for 1975 was 25.9, for 1976 22.2, and for the first 9 months of 1977, the average was 20.5. It seems unlikely that 1978 will be quieter geomagnetically than 1976 or 1977. Using $aa = 22.2$, the annual sunspot number at the forthcoming maximum epoch will be 206 ± 42 , that is, in the range 160–250, with a 66% probability. If the aa index for 1977 turns out to be lower than in 1976, a value of, say $aa = 20.0$ will still imply a predicted sunspot number of about 185 ± 40 .

As regards forecasting the next maximum epoch, Brunner (see ref. 4) showed a good inverse correlation between rise time of a solar cycle and the amplitude. Figure 2 shows the annual maximum sunspot number plotted against the time difference

Fig. 2 Annual sunspot no. at maximum epoch plotted against period of rise.

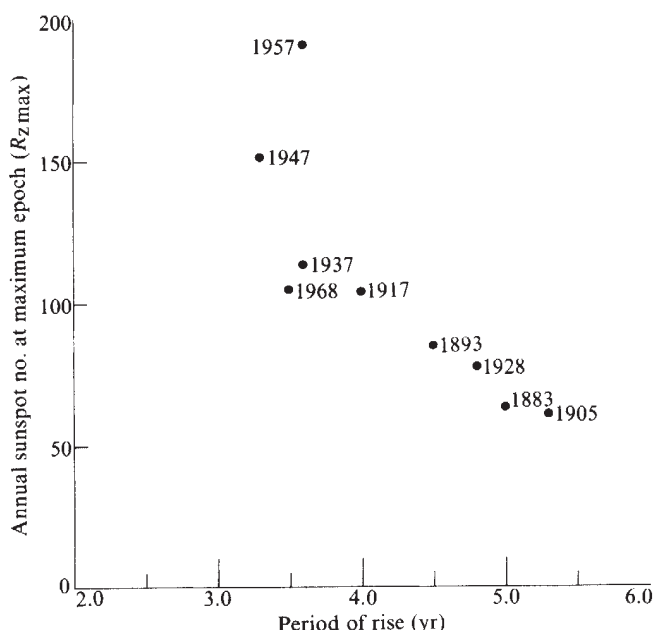


Table 1 Annual sunspot values for cycles 12–21

Solar cycle	Year of $aa_{\min}$	Annual $aa_{\min}$	Year of $R_{z\min}$	Annual $R_{z\min}$	Year of $R_{z\max}$	Annual $R_{z\max}$	Period of rise (yr)
12	1879	7.0	1878	3.4	1883	63.7	5.0
13	1890	10.8	1889	6.3	1893	85.1	4.5
14	1901	6.1	1901	2.7	1905	63.5	5.3
15	1913	8.7	1913	1.4	1917	103.9	4.0
16	1924	10.2	1923	5.8	1928	77.8	4.8
17	1934	13.4	1933	5.7	1937	114.4	3.6
18	1945	16.4	1944	9.6	1947	151.6	3.3
19	1954	17.3	1954	4.4	1957	190.2	3.6
20	1965	14.1	1964	10.2	1968	105.0	3.5
21	1976	22.2*	1976	12.6	—	—	—

*This is probably the minimum but the value for 1977 could be comparable or slightly less.

between maximum epoch and the preceding minimum epoch. The data for the past 100 yr show that the higher the amplitude of the maximum the shorter the rise time from the preceding minimum. The recent minimum was in July 1976 and if the next maximum is 160–250, the time of maximum will be 3.0–3.5 yr later than July 1976, or some time near the end of 1979.

It is interesting that the minimum monthly averages of aa indices do not correlate so well with the maximum sunspot number. The correlation is only +0.81, in contrast to +0.89 obtained by using the minimum annual averages. Also, sunspot numbers in years of minima do not correlate well with sunspot numbers at the following maxima—the correlation is only +0.27. However, a multiple correlation analysis where the dependent variable y is maximum sunspot number $R_{\max}$ and the independent variables are x as the minimum annual aa and z as the minimum sunspot number $R_{\min}$, both preceding $R_{\max}$, gives the following results for the correlations: $r_{yx} = +0.89$, $r_{yz} = +0.27$, $r_{xz} = +0.63$, $r_{yxz} = +0.97$, $r_{yzx} = -0.86$.

Thus, the correlation between maximum sunspot number and aa indices improves considerably and the correlation between maximum and minimum sunspot numbers is now highly negative, implying that the quieter the Sun during sunspot minimum, the larger would be the activity that follows, a situation similar to a calm before the storm in open seas.

The regression equation with standard errors is:

$$y = (13.0 \pm 1.1)x - (7.4 \pm 1.5)z - (3.4 \pm 10.3)$$

Using $x = 22.2$ and $z = 12.6$ for 1976, the predicted y , that is, $R_{\max}$ for cycle 21 is 192 ± 33 . Thus, the expected level is about the same as the sunspot activity in 1957 (which was the highest maximum in the historical record) and would be in the range 160–225 with 66% probability, and in the range 125–260 with 95% probability.

This relationship implies that the geomagnetic activity during the quiet Sun years is a measure of the intensity of the processes in the Sun which later show up as the number of surface spots at the maximum of the cyclical variation. It also implies that the sunspot number near minimum epoch is a poorer measure of the intensity to be expected for the growing cycle than the complex solar activity which causes geomagnetic activity. In fact, the partial correlation in this case is negative, which is surprising.

Let us compare our prediction for cycle 21 with other recent ones. Ohl¹ predicted that the maximum value of the annual average Wolf's number for cycle no. 21 would be 140–180, and Sargent² predicted a number > 150 . Using spectral analysis, Gleissberg⁵ predicted a mean sunspot number between 56 and 96, Cole⁶ predicted a number about 60 and Cohen and Lintz⁷ predicted a number about 50. In contrast, Smith⁸ predicts a number exceeding 150 and Hill⁹ one of at least 130 and possibly as high as over 200. Thus, whereas some of these roughly agree with our prediction, others predict very low solar activity. What actually happens remains to be seen.

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Solar plagues and the vorticity of the Earth's atmosphere

THREE superimposed epoch (SPE) analyses of the vorticity area index (VAI) at 500 mbar were carried out using the following definitions of the zero days: (1) the central meridian passage (CMP) of very active solar plagues (Fig. 1a); (2) the occurrence of peak values of the 10.7 cm solar radio flux (Fig. 1b); and (3) the CMP of active solar plagues also accompanied at CMP by sharp rises in 10.7 cm solar radio noise (Fig. 1c). All three SPE analyses show a sustained rise in VAI several days before the zero day continuing through the zero day, followed by a sustained minimum in VAI several days after the zero day. All three criteria for the zero date give results that are more clear cut than our earlier ones involving simply the occurrence of large solar flares, irrespective of their location on the apparent solar disk. The results lead us to conclude that the location on the solar disk of very active plagues plays an important part in determining their meteorological influence. We speculate here that the initial rise in VAI is caused by enhanced electromagnetic radiation associated with the solar activity, and that the decrease some days later is the result of the geomagnetic storm particle emission that generally follows the zero date.

In earlier work¹ evidence was found for the effects of large solar flares on the atmospheric circulation at 500 mbar, as shown by the VAI. The VAI is a measure of the area of the Northern Hemisphere in which the positive (cyclonic) vorticity of the atmosphere is above certain thresholds. In further work (unpublished), we verified the above result using independent data². We also found that the flares which occurred near central

meridian (CM) on the solar disk were those which caused the bulk of the observed effects. We decided, therefore to examine systematically the VAI response to CMP dates of large, active solar plagues.

The activities of solar plagues from 1954 to 1974 have been evaluated³ and an activity index, the active region index (ARI), prepared based on the following characteristics of the plague: the size and complexity of the accompanying sunspots; the flare productivity; associated sudden ionospheric disturbances (SIDs); centimetric and metric radio flux emission from the plague. The method of computing the ARI is given in ref. 3. (The list of CMP dates and ARI values can be obtained from the authors.)

Values of the ARI range from 1 to 19. There are 92 plagues with 91 different CMP dates of index 10 or greater in the 20-yr period 1954–74. They are reasonably well isolated in time. As the daily VAI values comprise a rather noisy data set, we normalised the data by selecting the three maximum and three minimum days per month, based on the average VAI value for each day. The procedure for picking these days was to consider all days of the month as an array, then to select the three highest and lowest VAI values.

Figure 1a shows the first results (all seasons and all solar cycles have been amalgamated). Using superimposed epoch analysis, with CMP date of the plague defining the zero point, we have plotted the number of maximum VAI days minus the number of minimum days. From day -2 to day +1 the frequency of occurrence of a maximum VAI day is significantly greater than the frequency of a minimum day. At day +2 the maximum and minimum VAI curves have begun to reverse their trends. The times when the maximum and minimum VAI frequencies are significantly different, and when geomagnetic activity is significantly above background are indicated.

Because we do not know which mechanism is involved in the plague–VAI relationship, we next used a solar radiation measurement to define the zero day. Solar radio flux data are available on a daily basis from 1947 to the present from measurements in the 10.7 cm band by Covington at Ottawa. This flux correlates well with other measures such as sunspot number and X-ray flux, and is a good basic solar activity index. To establish zero dates we selected as the solar parameter the three days per month on which the Ottawa flux was greatest. For a few months Ottawa data were incomplete, and sunspot numbers were used in their place. Figure 1b shows the result. The general agreement between Fig. 1a and 1b is not surprising. The 10.7 cm flux tends to maximise near times of CMP of large plagues, so Fig. 1a includes many of the same zero days as Fig. 1b. However, the data points in Fig. 1b are much more numerous, as we included every month from 1946 to 1975 regardless of position in the solar cycle; also they cover a longer time period.

Because both plague CMP and high values of centimetric flux seem to be associated with high values of VAI, we combined the two parameters and arrived at another set of zero dates. The new zero date is defined as a day of maximum centimetric flux, which is also the day of CMP (± 1) of a plague of any index from the Dodson list: Fig. 1c shows the result. The maximum VAI days are significantly more frequent than minimum days from day -7 to day +1, and the reverse is true for days +3 to +8. This suggests that any plague of greater than average activity, if it produces a maximum in 10.7 cm flux at CMP, is associated with an increased number of maximum VAI days as soon as the plague crosses the east limb. This increased VAI level maintains itself until the geomagnetic activity starts, and then the VAI reverses and remains at a low level for about a week. We consider Fig. 1c to be the most significant result, as it is based on two solar criteria and as the resulting effects on the VAI occupy sustained periods of time.

This suggests that solar data can be important in weather forecasting. Solar forecasters can now exhibit some skill in predicting the effect of plagues on the centimetric flux and geomagnetic activity by using the 27-d recurrence tendency

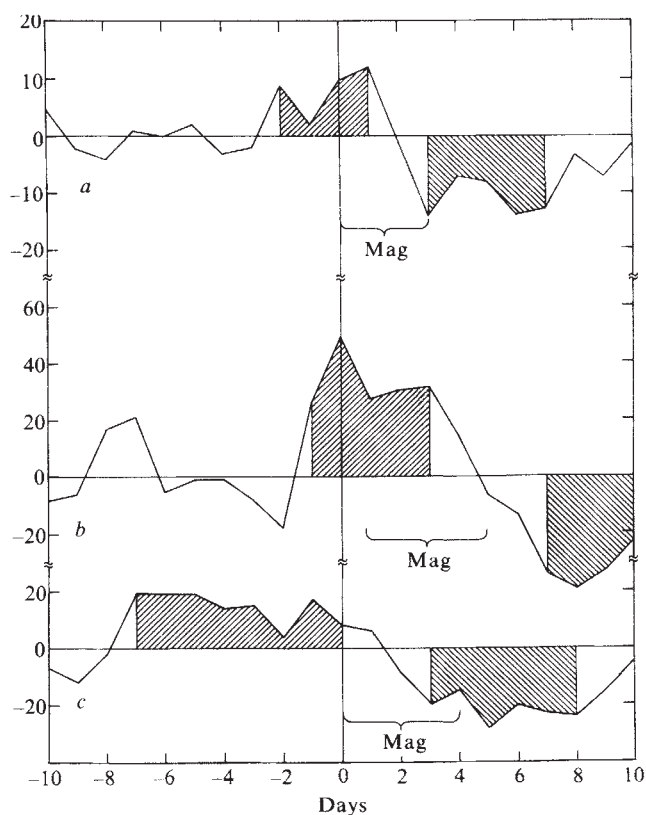


Fig. 1 *a*, The number of maximum minus the number of minimum VAI days before and after zero days 1955–74. Zero days are defined as the days of central meridian passage of a plague of activity index ≥ 10 on the Dodson–Hedeman list. *Mag* indicates the days when geomagnetic activity was significantly increased ($N=91$). The cross-hatched areas indicate when the maximum and minimum VAI frequencies are significantly different. *b*, As for *a* except zero days are the three maximum days per month of the 10.7-cm solar radio flux, 1946–75 ($N=1,088$). *c*, As for *a* except zero days defined by two criteria: zero days are days of maximum 10.7-cm flux; zero days are also days of CMP (± 1) of a plague of any activity index from the Dodson–Hedeman list, 1955–74 ($N=278$).

and by assessing the appearance and activity of each plague as it rises over the east limb. This information, combined with standard meteorological information, could perhaps improve the current long-range weather forecasts. As forecasting skill is quite low, beyond about five days, an experiment to determine the feasibility of adding information about vorticity based on this plague relationship seems worthwhile. One advantage of the results shown in Fig. 1c is that they are almost as clear in years of low solar activity as in more active years, in contrast to the flare results.

After completing this study, a new list of active plagues, with activity index ≥ 10 , was drawn up (by E.R.H.) for solar cycle 18. This list contains 53 plagues, with CMP dates from 1948 to 1951. Analysis of this list showed a large excess in the number of VAI maximum days on days -1 and 0, and a large excess in minimum VAI days on days +6 to +8. Although the data going into this list were not as reliable as the data for cycles 19 and 20, the agreement is rather convincing.

In previous work we had found the following relationships between solar flares and the VAI. First, the VAI tends to be at unusually high values around the time of large solar flares. After the geomagnetic storm which frequently follows the flare, the VAI is unusually depressed for a few days. The departures are 5–10% above and below background. Second, there is a weak seasonal trend in the effects, with summer months somewhat less prone to show the effects. Third, the effects are seen in all three of the most recent solar maxima (cycles 18, 19

Table 1 Number of maximum and minimum VAI days from day -1 to +1 and from day +5 to +10

	Fig. 1a		Fig. 1b		Fig. 1c	
	Days -1 to +1	Days +5 to +10	Days -1 to +1	Days +5 to +10	Days -1 to +1	Days +5 to +10
No. of max. days	37 (25)	37 (58)	373 (322)	551 (628)	96 (80)	114 (170)
No. of min. days	13 (25)	79 (58)	271 (322)	704 (628)	65 (80)	227 (170)

The three main parts of the table correspond to the three parts of Fig. 1. Random expected numbers are shown in parentheses.

and 20). The effects are strongest when flares occur frequently rather than in isolation.

The results described here, using plage CMP days as zero days, generally agree with those obtained with the flares. However, the plage dates seem to organise the VAI data slightly better than the flare dates. Flares may tend to occur at various locations on the solar disk at about the same time, thus tending to obscure the results.

Figure 1 shows the times when we determined that the numbers of maximum and minimum VAI days are significantly different. Another test of the significance of the maximum VAI-minimum VAI difference seems advisable. The most consistent pattern shown in the three graphs is the excess of maximum VAI days around day zero and the excess of minimum days from day +5 to day +10, and Table 1 shows this difference. This table can be tested for significance, by assuming that the values being grouped together are statistically independent, that there is no serial correlation in the data. As there is, in fact, serial correlation in weather data, this assumption is not valid, so we corrected for this fact in a very conservative fashion. If the three maximum days and three minimum days per month were always consecutive, the proper procedure would be to divide the number in Table 1 by three before testing. Although the amount of serial correlation is by no means that strong, we did, in fact, divide all numbers by three, and then carried out a χ^2 significance test.

Even with this very conservative approach, we found that, for each of the three parts of the contingency table, the probability of obtaining from a random set a distribution as highly skewed as shown is $<0.5\%$. Applying the same test to the increase in geomagnetic activity which follows the zero day in all three cases gives results which are also significant, but at a somewhat lower level.

We conclude that plages are perhaps clearer than flares as solar signals related to Sun-weather effects. This is partly due to the statistical fact that plage CMP dates are generally well separated in time. However, another reason may be that the plage is a stronger energy source at the Earth. A typical flare can last from a few minutes to a few hours, while a plage is a continuous source of energy. It seems likely that the increase in VAI at and before zero day is an electromagnetic radiation effect. The decrease in VAI a few days after CMP is, we think, more likely to be caused by the low energy particle radiation that produces geomagnetic storms.

We have discussed the importance of the differences in solar signals; the explanation of any Sun-weather effect must probably also rest on the condition of the Earth's atmosphere before the solar signal is felt. Hines and Halevy⁴ found that the effect of solar sector boundaries on the VAI was enhanced if the preceding VAI data showed a high noise level. An analogous finding from our study is that the effects we found seem to be enhanced if the VAI is elevated for a few days before the solar effects are felt.

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Fragmentation requirements for detonating thermal explosions

THE contact of a hot with a cold liquid can develop into a rapid thermal interaction in which a significant fraction (approaching the thermodynamic limits) of the thermal energy in the hot liquid is converted into mechanical work. These so-called 'thermal explosions' are of interest to several industries, such as paper, metal, LNG and nuclear power reactor. One approach towards understanding thermal explosions has been proposed by Board *et al.*¹ They used ideas of classical detonation theory in conjunction with a mechanism of pressure wave-induced fragmentation of liquid fuel drops premixed (coarsely) with the liquid coolant, the quantitative aspects of which they based on existing measurements in gas-water systems. We report here our studies of fragmentation of high surface tension liquid drops (mercury) in a liquid medium (water) in which we found that the fragmentation behaviour is considerably different from that in the gas-water system.

Extremely fine and rapid fragmentation of the hot liquid is necessary for large scale energetic thermal interaction. The fine particles provide the high heat transfer rates, and the short time scales provide coherent behaviour. Clearly, the requirements for these 'length' and 'time' scale are interdependent and their quantification must be accomplished by a thermal explosion model that takes into account both the motion and thermal states evolution of the system as a whole (including constraints). On the other hand, these requirements must be examined in the context of the physical mechanisms that produce fragmentation to assess the self-consistency and to show how the explosion model could be applied to a particular situation. The current state of explosion modelling can be represented by the drop capture and superheat model (DCSM) of Henry and Fauske² and the detonation model (DM) of

Board *et al.*¹ The fundamental difference between the two models is in the fragmentation mechanism invoked. According to the DCSM, which is related to the previously proposed spontaneous nucleation criterion, fragmentation due to 'explosive boiling' is a necessary condition for thermal explosions.^{3,4} According to the DM, which is related to the detonation mechanism of chemical explosions, fragmentation is due to hydrodynamic instabilities. Such instabilities are tied to the passage of pressure waves and the resulting differential accelerations of the premixture components. The important implication is that, given a particular coarse premixture, the DCSM leads to certain well defined temperature criteria while the DM leads to pressure pulse trigger requirements. (The relative merits of these models, other explosion models and a summary of the available experimental evidence, are discussed in ref. 5.) Here we focus our attention on fragmentation by hydrodynamic instabilities.

The problem of acceleration-induced fragmentation has been studied extensively for gas-liquid systems (liquid drops in

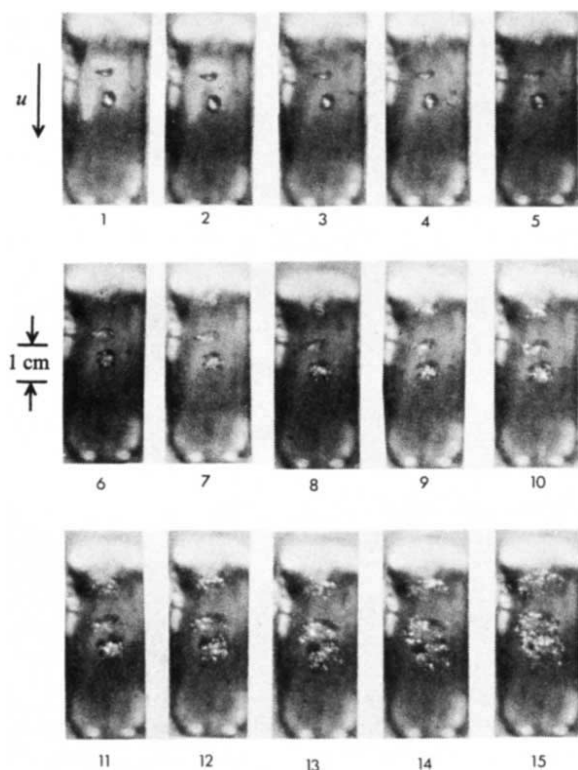


Fig. 1 Photographic sequence showing the mercury drop response to the shock-induced flow in water ($Bo = 67$). The shock impacts the drop sometime between frame 1 and 2. Time separation between each frame 1–10 = 220 μ s; between each frame 10–15 = 440 μ s.

a gas continuum) in connection with the erosion in supersonic flight due to raindrop impact and the combustion of atomised chemical fuels. Experimental evidence suggests that there are two breakup regimes. The Bond number ($Bo = \rho_d a_d R_d^2 / \sigma$, where a_d , ρ_d , σ , R_d , are the acceleration, density, surface tension and radius of the drop, respectively) which represents the ratio of acceleration forces to surface tension forces, provides the criterion for characterising drop response. For $Bo > Bo_{cr}$, and $Bo_{cr} \sim 10^4$ the response is dominated by acceleration effects, which cause Taylor instabilities on the windward surface of the drop and shattering exponential in time⁶. The drop

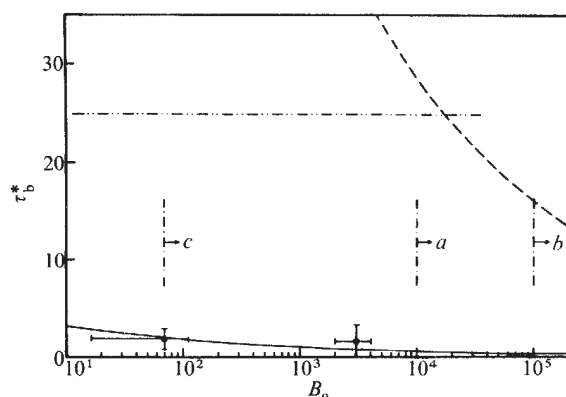


Fig. 2 Nondimensional breakup time plotted against Bond number. ---, Equation (1); · · · —, equation (2); —, equation (4) with constant of proportionality = 1.5; ●, present data; a, experimental exponential breakup threshold of ref. 6; b, theoretical exponential breakup threshold of ref. 7; c, instability limit of present work.

breakup time, $\tau_b^* = t_b u_g / R_d$ (where u_g is the gas velocity), is given by

$$\tau_b^* = 75 Bo^{-1/4} \epsilon^{1/2} \quad (1)$$

where ϵ is the liquid-to-gas density ratio. (Depending on the interpretation of the experimental results, the constant of proportionality may be 44 instead of 75. A constant of 22 was found to quantify the onset of instability⁶.) These results agree with the theory of Harper *et al.*⁷ who predict $Bo_{cr} \approx 10^5$ and $\tau_b^* \sim 0$ ($\epsilon^{1/2}$). For $Bo < Bo_{cr}$ aerodynamic effects dominate, the drop deformation is algebraic in time and a boundary layer stripping mode of breakup^{8,11} gives, for mercury–water,

$$\tau_b^* = 25-45 \quad (2)$$

These results were used in the original presentation¹ and in subsequent criticisms^{9,10} of the detonation model. Recognising the potential for a fundamentally different behaviour in a high interfacial tension liquid–liquid system, we will examine these fragmentation requirements more carefully.

We have investigated drop fragmentation characteristics in liquid–liquid (water–mercury) systems using a hydrodynamic shock tube. The driven section of the tube was filled with water and the driver section pressurised with nitrogen (up to 500 bar). Uniform flows of 2 ms duration were obtained. The response of falling mercury drops to the hydrodynamic shock was photographed at rates up to 6,500 frames s^{-1} . We have covered the following ranges of experimental parameters: drop diameters 2–6 mm and shock pressure ratios (driven section always at 1 bar) 60–340. This implies a nominal Bond number range of 67–3,000. The drag coefficient, which is needed in the determination of the Bond number, was estimated from the drop acceleration data as ~ 2 during the free fall of the drops and was within experimental error from the expected value⁶ of 2.5 following the shock impact.

The results reveal a different behaviour than that expected from previous work. In all cases, a fine and rapid fragmentation without any hydrodynamic deformation or boundary layer stripping was observed. Over the range investigated, the effect of the Bond number was to intensify rather than alter the phenomenon: that is, no threshold values were evident. A typical photographic sequence is shown in Fig. 1 for the smallest Bond number case. The drop is seen to 'blow-up' rather abruptly due to fragmentation. Fragmentation seems to take place simultaneously in all directions. This blow-up phenomenon is clearly evident in drop diameter against time plots and its inception was used to define the breakup time.

Our experimental results, including upper bound estimates on experimental uncertainties, are compared with the behaviour of gas-water systems^{6-8,11} in Fig. 2.

On the basis of the morphology of the fragmentation process in the water-mercury system, we propose that Taylor instabilities are responsible for breakup for any Bond number greater than the minimum investigated here (that is, 67). Support for this hypothesis comes from the instability growth times. The results of Bellman and Pennington¹² indicate that the instabilities grow (linearised analysis) as $\cosh(n\tau)$ where n , for the fastest growth, is given by

$$n^2 \sim \frac{(g(\rho_2 - \rho_1))^{3/2}}{\sigma^{1/2}(\rho_2 + \rho_1)} \quad (3)$$

This, with $\rho_1 \ll \rho_2$, leads to a time constant of

$$\tau^* \sim B_0^{-1/4} \epsilon^{1/2} \quad (4)$$

Figure 2 shows that this equation with a constant of proportionality 1.5 agrees well with our data. This also corresponds to one time constant in the asymptotic approximation (for high wave numbers) for the fastest growing instability given by Harper *et al.*⁷

$$\eta_{s \max} = 1 + 1/2 \gamma P_{N \max} \exp \left\{ \frac{0.62 B_0^{1/4}}{\epsilon^{1/2}} t^* \right\} \quad (5)$$

The experimental correlations^{6,8} from gas-water systems, on the other hand, indicate that nearly 14 time constants are required⁶ for the onset of instability and nearly 47 for complete breakup⁸. We are investigating this interesting discrepancy.

Based on these present experimental results, the pressure pulse trigger requirements for the DM are drastically reduced compared with the original estimates of Board *et al.*¹ Fragmentation, however, is only one aspect of the explosion process. The vapour production and pressure feedback mechanisms are also an integral part of the escalation and they too must be investigated in order to assess the conditions of applicability of the DM.

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Planar solar energy convertor and concentrator based on uranyl-doped glass

A MAJOR improvement of solar cell efficiency is described here which uses a fluorescent plane glass to convert and concentrate the ultraviolet (UV) and blue part of the solar spectrum, thus increasing the possibilities of photovoltaic solar energy conversion. An advantage of our device is that the heat energy coming directly from the Sun will be dissipated over the

Table 1 Ratio of the short-circuit current obtained by concentrating the applied radiation (mercury lamp) by a uranyl-doped glass to that of using an undoped glass

	wt%	UO ₂ ²⁺	In glass
	0.5	1.0	2.0
I/I_{blank}	3.10	3.00	2.54
I/I_{cell}	1.54	1.50	1.06

large area of the glass and only the energy in the visible part of the spectrum will reach the solar cell. Moreover, the excess between the absorbed and band-gap energy which is evolved as heat in the solar cell will be diminished by decreasing the difference in wavelength between the useful and excess energy, and thus the solar cell will be heated to a much lesser extent.

At present, silicon semiconductors have a key role in most types of solar energy conversion because of their opto-electronic properties, and the highly developed technology of their production. However, the cost of the cells is still too high to permit large-scale terrestrial use of solar to electrical energy conversion. One technique for improving the performance and

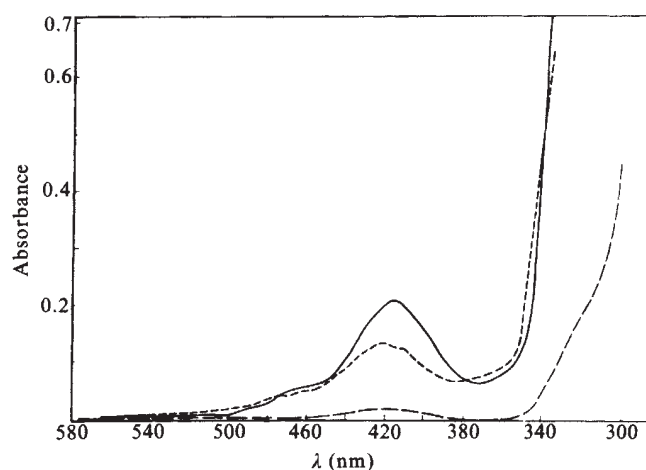


Fig. 1 Absorption spectrum of UO₂²⁺ in phosphate glass. 1 wt% (—); 0.5 wt% (---); 0.1 wt% (-.-).

reducing the cost of the cell power is to concentrate the solar flux, thus increasing the conversion efficiency and greatly reducing the necessary solar cell area.

It has been suggested that cheaper and lighter solar cell power supplies could be made by focusing the Sun's light onto the cells. Most of the power supply's cost and weight could then be reduced. The existing collectors concentrate not only the visible part of the spectrum that is utilised by the solar cells which have their maximum sensitivity around 800 nm, but also concentrate the heat energy, which has a significant lowering effect on the conversion efficiency¹.

Luminescent solar collectors have been suggested by Weber and Lambe² in which the luminescent material is neodymium doped laser glass or rhodamine-6-G. Their calculations link the trapping of the radiation with the refractive index of the collector. Following this idea we use a medium which strongly absorbs the UV and blue part of the solar spectrum and emits longer wavelengths. The emitted radiation, contrary to the exciting radiation, is not absorbed by the glass and is collected at the edge as most of the light is emitted at angles more grazing to the surface than the initial angle of total internal reflection. The trapped light propagates by successive reflections to the narrow edges where it is finally allowed to escape at one edge. A similar effect is known in fibre optic waveguides.

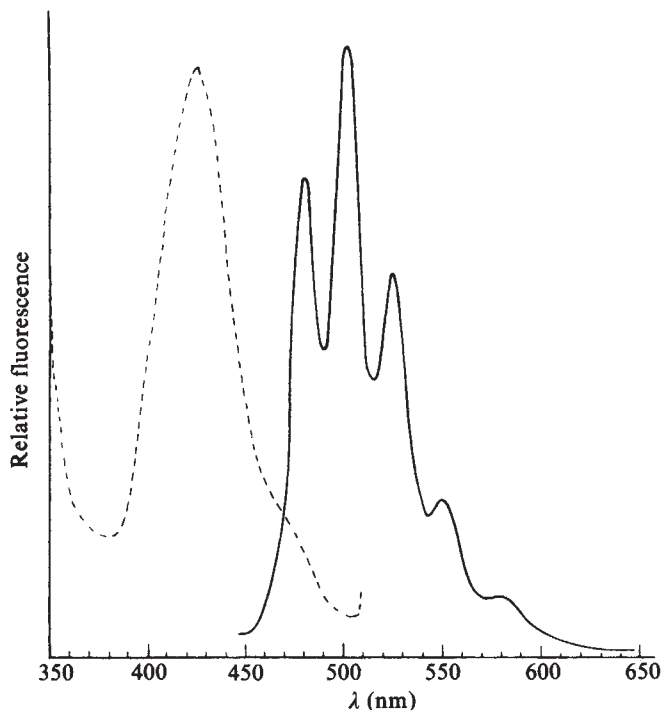
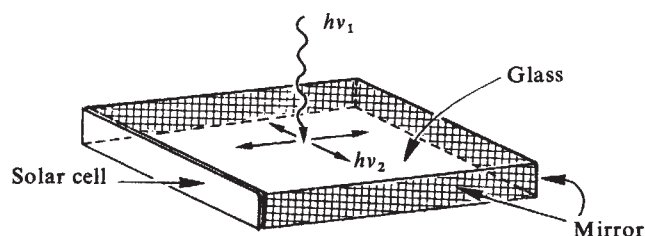


Fig. 2 Emission spectrum of 1 wt% UO_2^{2+} in phosphate glass. Excitation $\lambda_{\text{Em}} = 507 \text{ nm}$ (---); emission $\lambda_{\text{Ex}} = 423 \text{ nm}$ (—).

We use here uranyl-doped glass as a planar collector. The reasons for this choice are (1) the absorption of the uranyl ion is five orders of magnitude higher than Nd^{3+} ; thus, the solar light is totally absorbed with small doping levels of UO_2^{2+} ; (2) the uranyl ion absorbs in the part of the spectrum to which the silicon cells are not sensitive; thus, the uranyl ion can utilise energy that would otherwise be lost; (3) uranyl fluoresces with high quantum efficiency at room temperature at longer wavelengths at which the sensitivity of the silicon cell is higher than to the absorbed radiation; (4) the geometrical arrangement proposed permits the use of only the visible part of the spectrum and the unwanted heat is dissipated by the large area of the glass; (5) nonradioactive uranyl is abundant as it is a byproduct of isotope separation of radioactive ^{235}U and is thus inexpensive.

The absorption spectrum of the uranyl ion in phosphate glass as studied by Lieblich-Sofer, Reisfeld and Jørgensen³ is presented in Fig. 1. The origin of this spectrum^{4,5} arises from the electronic transfer where the electron is excited from the molecular orbitals consisting almost exclusively of 2p oxygen orbitals in the LCAO model to the empty shell of the uranium atom. Excitation of uranyl in these bands results in a strong visible fluorescence (Fig. 2) which has a quantum efficiency of about 30% at room temperature and a lifetime of around 300 μs (ref. 3).

Fig. 3 Schematic representation of the concentrator and convertor. Mirrors cover the underside and three edges of the converting glass.



When a rectangular slab of uranyl-doped glass is coupled to a silicon cell (see Fig. 3) and excited by a high-pressure mercury source, a considerable increase of current and voltage is achieved. This is shown in Table 1, which gives the ratio of short-circuit current of the cell obtained by the concentrated light by the collector to that of the current obtained from the cell connected to a blank undoped glass of this same geometry, I/I_{blank} , and the ratio of current obtained by the cell connected to the collector against the cell exposed to direct illumination, I/I_{cell} . At higher concentrations of uranyl the ratio is decreased as a result of fluorescence concentration quenching of uranyl. The values were obtained when the ratio of the glass area to the silicon cell was about 18. An additional increase can be expected by increasing the ratio of glass to cell areas.

Note that the result was obtained using only uranyl-doped glass and that the luminescence of uranyl does not fall in the spectral range of maximum sensitivity of silicon cells which is $\sim 800 \text{ nm}$. This difficulty can be avoided by energy transfer from the uranyl ion to an ion emitting closer to the maximum spectral sensitivity of silicon. Such energy transfer was carried out in our laboratory as a way of improving the laser characteristics of rare earth-doped glasses^{6,7}.

Because of the recent development in highly concentrated silicon cells which can receive a large number of photons per unit area without decreasing the fill factor the present development may be of practical use.

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Roll cell mantle convection under the Pacific plate

SPECTRAL analysis of bathymetry and heat flow data for selected regions of the Pacific has revealed anomalous patterns which trend in the direction of absolute motion of the Pacific plate. The bathymetry and heat flow anomalies have dominant wavelengths of the order 1,000 km, and amplitudes about 300 m and 10 mW m^{-2} respectively. We suggest here that these observations support the hypothesis of a roll cell form for small scale mantle convection as advanced by Richter and Parsons¹.

The Pacific plate (Fig. 1) was selected as a test region because it is fast moving, of large dimensions and its sense of motion has been reasonably constant for the past 40 Myr. Data used in this study include shiptrack bathymetry³, and heat flow and bathymetry pairs⁴. The data were treated as discretely sampled waveforms and analysed for their spectral characteristics using both the maximum entropy and fast Fourier transform techniques. The Burg algorithm⁵ was most useful in producing enhanced spectral peaks while the Fourier transform provided amplitude and phase information.

Raw bathymetry data and corresponding amplitude spectra for two representative shiptracks are shown in Fig. 2. Shiptrack AA' is transverse to the direction of motion of the Pacific plate while BB' roughly parallels the plate motion. Whereas the spectra for both tracks contain power at long wavelengths

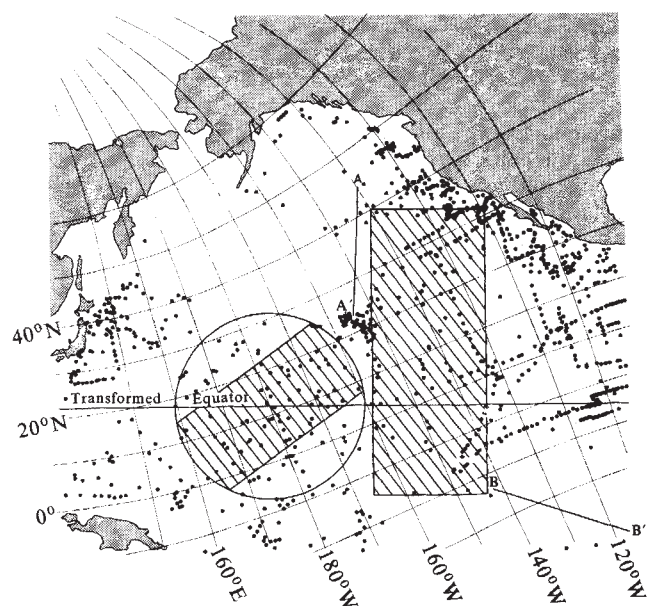


Fig. 1 The Pacific region transformed into a Mercator projection about the Pacific plate instantaneous rotation pole at 67°N, 59°W (ref. 2). Solid dots indicate heat flow sites. Analyses discussed in text were performed on data along tracks AA' and BB', and in shaded windows.

(frequency <0.5 cycles per 10^3 km) and also at short wavelengths (frequency >2.5 cycles per 10^3 km) the transverse spectrum AA' shows a significant peak in the frequency range 1.2–1.4 cycles per 10^3 km corresponding to wavelengths of about 800 km, which is absent on the parallel spectrum BB'. Of several shiptracks considered sufficiently long and correctly orientated for this study, all except one exhibit this pattern of behaviour in the 700–1,000 km wavelength range.

A similar analysis was performed on heat flow–site bathymetry pairs in selected spatial windows. After removing the mean values we projected the residuals on to profiles transverse and parallel to the plate motion and averaged the values over 0.5° intervals. In spite of the noise in the raw data, the maximum entropy spectra for the transverse profiles again show peaks in the frequency range of 1.2–1.4 cycles per 10^3 km while the spectra for the parallel profiles are subdued through the same frequencies. Fig. 3 shows a set of results for the transverse window outlined in Fig. 1.

Before performing further tests we examined the hypothesis that spectral power in the 700–1,000 km wavelength range might be produced by the cyclic juxtaposition of older and younger ocean floor due to offsets across fracture zones. For shiptracks we analysed theoretical bathymetry profiles generated from the established age–depth relation for the Pacific plate⁶. The spectra exhibited considerable power for wavelengths $>2,500$ km, but no spectral peak in the wavelengths range of concern. In the case of the spatial windows we corrected each bathymetry and heat flow value for the age effect and recomputed the spectra. Although the age corrections do modify the spectra (Fig. 3), they do not reduce the relative amplitude of the 700–1,000 km wavelength feature; thus we need explanations other than ocean floor age patterns for these peaks.

The azimuthal dependence of the 700–800 km wavelength feature was tested further in a circular region of the western Pacific (see Fig. 1), again using heat flow and bathymetry pairs. In this case a discrete waveform was generated by projecting all data points falling within a window 10° on either side of a projection axis onto the central axis and averaging the values over a 0.5° interval. Amplitude spectra for both heat flow and

elevation were determined for discrete axis azimuths ranging between 0 and 180° measured with respect to the instantaneous pole of motion. The azimuthal dependence of amplitude for the 720-km wavelength feature is shown in Fig. 4. Both heat flow and bathymetry spectra display relative amplitude maxima near 5° , a direction approximately transverse to the direction of plate motion, and relative amplitude minima in the direction parallel to plate motion. Furthermore, the similarity in the azimuthal dependence of the two spectra suggests that the bathymetry anomaly has a thermal origin.

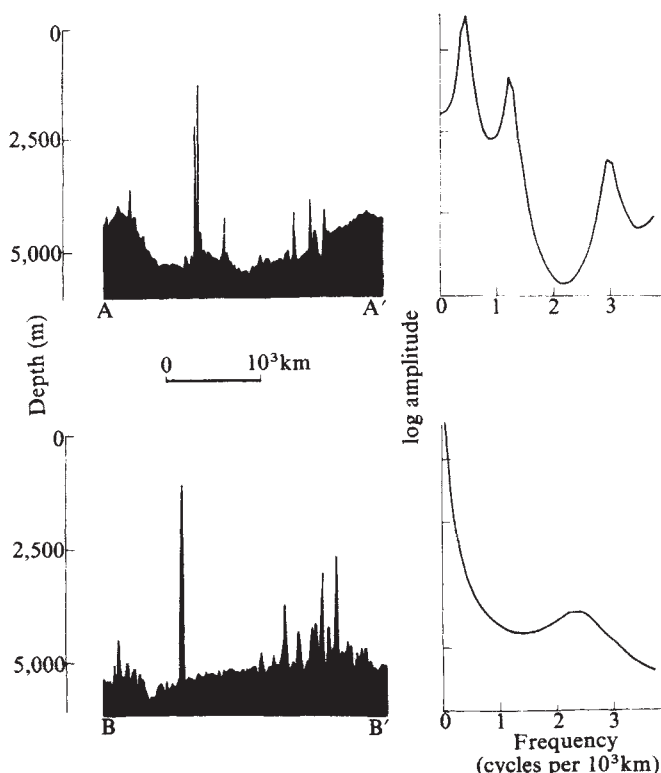


Fig. 2 Bathymetry and corresponding maximum entropy amplitude spectra for shiptracks AA' and BB'. Amplitude scale divisions are decade increments determined using the Burg algorithm. The 800-km wavelength peak for AA' has amplitude approximately 300 m.

We now consider the implications of our bathymetry and heat flow spectral analysis for the scale of mantle convection under the Pacific plate.

A system in which convection takes place in the upper mantle on two distinct horizontal length scales has been proposed by Richter and Parsons¹. Large scale convection consists of lithospheric plates themselves and the return flow in the mantle necessary to conserve mass. Such convection has a horizontal scale of plate dimensions typically 5,000 but as large as 10,000 km. Small scale convection is envisaged as a variant of Rayleigh–Benard convection operating immediately below the plates. Richter and Parsons¹ suggest that this small scale convection extends from the base of the plates down to the 650-km seismic discontinuity, and hence the characteristic horizontal scale would be some 550 km or an order of magnitude less than that of large scale convection. An important property of the system is the interaction of the two flows, in which the shearing action of moving plates transforms the equidimensional small convection cells into long tubular roll cells with their axes parallel to the direction of the overall plate motion. For fast moving plates such as the Pacific plate the roll cell alignment may be accomplished in a geologically short time whereas under slow moving plates the convective platform is

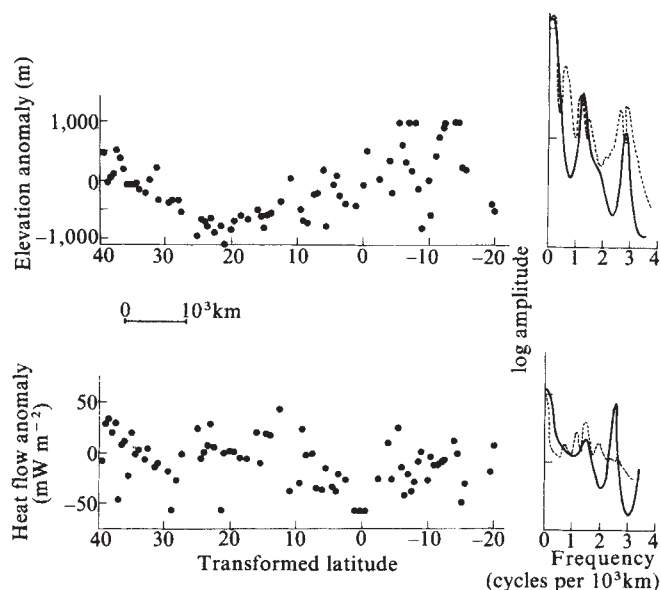


Fig. 3 Elevation and heat flow anomalies with their maximum entropy amplitude spectra for data within the transverse rectangular window. Site latitudes are measured in the transformed Mercator projection of Fig. 1. Solid lines represent spectra for raw data; broken lines for age corrected data. Amplitudes of the 800-km wavelength peak are about 300 m and 10 mW m^{-2} respectively.

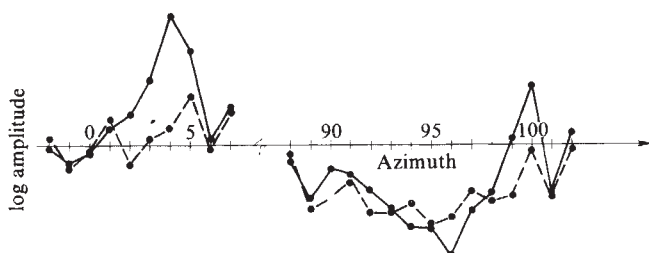
likely to be irregularly spaced upwelling and downwelling spouts.

Direct observation of small scale mantle convection is difficult because plates themselves act as shields in masking temperature variations below them, this masking being aggravated by any relative motion between the plate and the convective planform⁷. In this regard the relative stationarity of roll cell axes will greatly enhance the possibility of their detection.

Before this study, two other sets of orientated anomalies in the Pacific region have been linked to the existence of roll cells. Selected harmonics of the global free air gravity field show a distinct trend in the direction of plate motion⁸ as do residual depth anomalies⁹. However, if these features result from roll cell convection, their respective transverse wavelengths of 2,000 and 4,000 km imply convection to significantly greater depths in the mantle than was suggested by Richter and Parsons.

Our preliminary combined heat flow and bathymetry result supports the concept of small scale convection under the Pacific plate while suggesting a lateral scale for the convection cell that previously has gone unnoticed. A roll cell system having axes separated by about 800 km and parallel to the instantaneous

Fig. 4 Azimuthal test for heat flow (solid line) and bathymetry (dashed line) data in the circular window of the central Pacific (Fig. 1), $\lambda = 720 \text{ km}$.



motion of the Pacific plate would produce effects consistent with our observations. Other possible sources for the anomalies such as sediment thickness variations or spacing of sea mounts, fracture zones and other obvious physiographic features have been examined but none produce significant amplitude peaks in the wavelength range of concern. Further investigations involving analysis of wavelength dependent gravity and residual depth anomaly correlations as discussed by McKenzie¹⁰ and the acquisition of closely spaced precision heat flow and bathymetry data along selected shiptracks seem warranted.

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Behaviour of rare earth elements during submarine weathering of tholeiitic basalt

DREDGED oceanic basalts and those sampled by deep-ocean drilling have commonly undergone some weathering with subsequent changes in their chemical composition^{1–7}. The rare-earth elements (REE) La–Lu are generally considered to be unaffected by weathering and are thus often used to characterise variations in basalt and magma composition. When considered relative to chondritic abundances⁸, the overall REE distribution is a critical diagnostic tool. In particular, the relative depletion of light-REE is used to discriminate between magma types. We present here new findings which show that REE are affected by low-temperature weathering processes. In particular, the light-REE show enrichments which may result in a basalt with a primary light-REE depleted pattern showing a flat or enriched light-REE pattern.

The samples studied were selected from basalts recovered during a dredging transect at 23°N at distances of 0–725 km from the Mid-Atlantic Ridge. These samples represent a time sequence of basalts erupted at a single point on the ridge axis over the past 57 Myr. Discussions concerning the mobility of the major elements and some trace elements for these samples during low-temperature weathering processes have been presented elsewhere^{5,6}. Our discussion is restricted to basalts dredged from near the median valley (samples <5 Myr). The samples analysed were: the most fractionated and least fractionated unaltered tholeiitic basalt from the median valley at 23°N (0 Myr); the crystalline interiors of basalts of 2.5, 4.5 and 5.0 Myr; the palagonitised rinds of the 4.5 and 5.0 Myr basalts (age dates are based on sample position relative to magnetic anomalies). The REE, H_2O^+ and $\text{Fe}_2\text{O}_3/\text{FeO}$ data are presented in Table 1.

The 0 Myr fractionated and unfractionated basalts represent pristine tholeiitic compositions erupted at the spreading centre

Table 1 REE* analyses of fresh basalts and weathered pillow basalt interiors and margins from 23°N

Age (Myr)	0 Ufr.	0 Fr.	2.5 Int.	4.5 Int.	4.5 Marg.	5.0 Int.	5.0 Marg.	BCR-1 Found	BCR-1 Flanagan ¹⁷
La	1.67	3.75	4.46	4.50	30.08	6.23	16.35	24.60	26.00
Ce	5.86	13.68	15.82	14.65	43.97	16.58	24.78	53.60	53.90
Nd	5.33	11.42	12.96	12.21	34.48	12.28	19.70	26.60	29.00
Sm	2.23	4.23	4.67	4.83	9.37	4.27	5.30	6.10	6.60
Eu	0.90	1.52	1.58	1.60	2.58	1.32	1.63	1.88	1.94
Tb	0.56	0.95	1.08	0.96	1.58	0.75	1.06	1.10	1.00
Ho	0.83	1.38	1.42	1.47	2.33	1.14	1.49	1.18	1.20
Yb	2.02	3.82	3.73	4.15	6.25	3.09	4.14	3.32	3.36
Lu	0.34	0.62	0.63	0.68	0.99	0.52	0.72	0.53	0.53
H ₂ O ⁺	0.11	0.20	0.51	2.03	7.37	2.37	11.14	—	—
Fe ₂ O ₃ /FeO	0.18	0.22	0.76	2.08	27.20	3.37	17.16	—	—

Ufr, unfractionated; Fr, fractionated; Int., interior; Marg., margins.

* Analyses by INAA of rock powders, all analyses are within $\pm 5\%$ with the exception of Ho $\pm 10\%$.

at 23°N; both samples show low H₂O⁺ and Fe₂O₃/FeO. The extent of weathering of the older basalt interiors is reflected by the H₂O⁺ (0.5–2.3 weight %) and Fe₂O₃/FeO (0.76–3.37). Both the 4.5 and 5.0 Myr basalt rinds are completely palagonitised. The high Fe₂O₃/FeO for the 4.5 Myr basalt probably reflects incorporation of ferromanganese deposit on the exposed portion of the pillow.

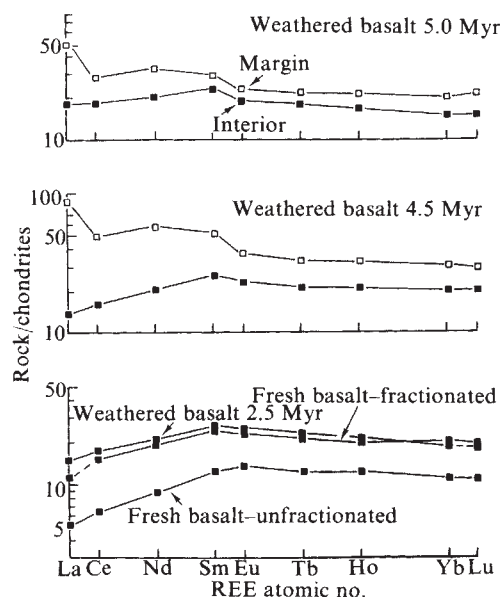
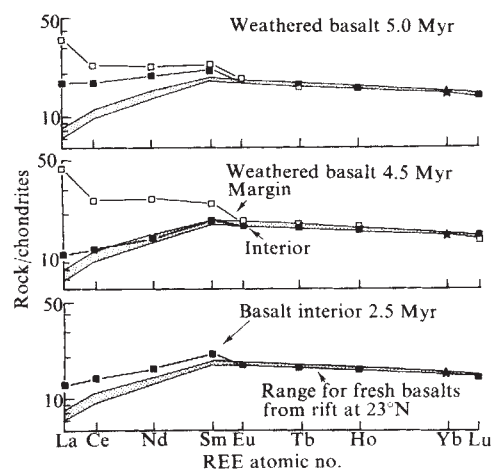


Fig. 1 Chondrite normalised REE profiles for fresh (unfractionated and fractionated) pillow interiors, weathered crystalline pillow interiors and palagonitised pillow margins from the ridge axis and ridge transect at 23°N.

The REE data are presented in Fig. 1. The behaviour of the REE during palagonitisation of the glassy-rinds during the low-temperature weathering process is most marked by the variation in abundance between the margins and interiors of the 4.5 and 5.0 Myr basalts. Both samples show a very significant La enrichment and a less pronounced enrichment in Ce, Nd and Sm. The ratios of the REE Eu–Lu remain constant between margin and interior indicating that there has been no selective enrichment of the heavy-REE. Thus, an element such as Yb, for which one may obtain good analytical data, may be

used to normalise an altered REE pattern to a primary pattern. This procedure may be applied in Fig. 1 for the 4.0 and 5.0 Myr pillows by superimposing the heavy-REE pattern of the pillow margin over that of its interior. This normalisation procedure confirms the extensive enrichment of La during palagonitisation (enrichment factors of 4.5–5.5 are observed); less pronounced enrichments for Ce (1.5–2.2), Nd (1.4–2.0) and Sm (1.3–1.5) are also observed. Constant-volume calculations in conjunction with trace element data have established that the older basalts dredged on the 23°N transect are equivalent to the 0 Myr basalts erupted at the ridge axis in terms of immobile trace elements such as Zr, Y and Ti; observed compositional ranges are: Zr, 60–100 p.p.m.; Y, 20–30 p.p.m.; TiO₂, 0.9–1.5 weight %. Therefore, these basalts are not considered to be large-ion-lithophile (LIL) element enriched magmas. A logical progression to the normalisation procedure is to assume that the REE abundance of the unweathered (0 Myr) fractionated and unfractionated tholeiitic basalts represents the complete range in REE for 'pristine' tholeiitic melts erupted at the spreading centre at 23°N. Thus, the abundance observed in the weathered basalt interiors and the palagonitised rinds may be normalised to an average Yb value for the unweathered basalts. The results of such a normalisation procedure are

Fig. 2 Chondrite normalised REE profiles for weathered pillow basalt interiors and margins after normalisation to constant Yb for fresh basalts from the ridge axis at 23°N. (The shaded region represents the range in abundance for the fresh basalts at the ridge axis. Departures from this region reflect element mobility during weathering.)



shown in Fig. 2. Any element falling within the shaded area is representative of the fresh tholeiite composition at 23°N, and departures from this pattern, in particular selected departures for certain elements, reflect the effect of seawater weathering. The remarkable constancy of the heavy-REE favour our argument for such a normalisation procedure. This procedure emphasises the conclusions outlined for the palagonitised rinds. The less altered ($\text{Fe}_2\text{O}_3/\text{FeO}$; 0.76 and 2.08) 2.5 and 4.5 Myr basalt interiors show only minor departures in the light-REE. However, the most altered crystalline interior, the 5 Myr basalt ($\text{Fe}_2\text{O}_3/\text{FeO}$, 3.37), shows a significant increase in La-Sm resulting in a REE profile similar to that observed for LIL element enriched tholeiites from the sea floor⁹⁻¹².

Figure 3 shows the chondrite normalised REE profiles for seawater $\times 10^7$ (refs 13, 14). Seawater displays a decreasing enrichment with atomic number relative to chondrites for the

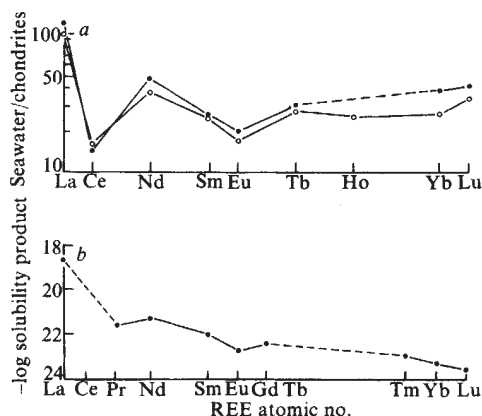


Fig. 3 Chondrite normalised REE profiles for seawater $\times 10^7$ (refs 13, 14). a (●), see ref. 14; (○), see ref. 13. Solubility product of the REE hydroxides¹⁸; b, taken from Glasby¹⁵.

light-REE and a relatively constant heavy-REE pattern. Seawater also displays a depletion in Ce; this has been attributed to the oxidation of Ce to the tetravalent state and its subsequent fractionation from seawater¹³⁻¹⁶. Thus, one may infer that the light-REE enrichment during low-temperature weathering is a reflection of the abundance of the REE in seawater. Such an argument is possibly more convincing for the older palagonitised basalt-rinds which often show a Ce depletion. Figure 3 also shows the variation in solubility product for the trivalent REE hydroxides¹⁸; these data indicate that the hydrolysis potential of the REE decreases strongly to Sm but is relatively constant from Eu to Lu. Thus, the hydrolysis potential of the REE may be the control of the fractionation of light-REE from seawater during weathering of seafloor basalt.

We feel that our data demonstrate significant uptake of the REE La-Sm during seafloor basalt weathering processes at low temperature. This process is accelerated in the palagonitised rinds of pillow basalts, all of which seem to be completely altered by 4 Myr. Although the weathering effect is slower in the crystalline interiors of pillow basalts, we observe the same process of light-REE enrichment, resulting in a tholeiitic basalt with a primary light-REE depleted profile bearing a profile similar to light-REE enriched tholeiitic basalts. We suggest, therefore, that REE data for weathered seafloor basalts be treated with caution, and that petrological inferences based on the REE abundances of seafloor basalts be restricted to fresh glass samples.

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Ionian Sea sapropel distribution and late Quaternary palaeoceanography in the eastern Mediterranean

SAPROPELS, dark organic-rich mud layers observed in deep-sea cores of the eastern Mediterranean, indicate that oceanographic conditions in this almost completely land-locked sea were at certain times during the Pliocene and Quaternary significantly different from those observed at present. Regional correlation of Quaternary sections in piston cores collected during the past two decades has shown that these distinct grey-to-black layers, ranging from a few millimetres to as much as 50 cm or more thick, are deposits that for the most part cover laterally extensive areas¹. It has been suggested that sapropel formation is directly related to phases of stagnation^{2,3} which occurred synchronously over much of the eastern Mediterranean⁴. It was proposed that this lithofacies type was associated with well-developed water mass stratification that induced euxinic conditions on the sea floor^{5,6} and accumulated primarily during periods of climatic warming. Recent studies indicate that some sapropels also formed during cooling trends within interglacial episodes⁷. Interpretation of Mediterranean palaeoceanographic conditions during late Quaternary stagnation phases requires a precise determination of both the spatial distribution and the age of sapropels. Correlations are generally made by lithologic, microfossil and oxygen isotope methods. The core-to-core variations (due to hiatuses or repetition of section) require a framework of absolute age determination in order to identify specific sapropels reliably over broad regions⁸. The study reported here is an extension of lithofacies and chronostratigraphic (based on ¹⁴C dating) investigations made by the Smithsonian Institution, of numerous piston cores recovered between the Strait of Sicily in the central Mediterranean and the Nile Cone in the eastern Levantine Basin. The regional distribution of late Quaternary sapropels is outlined and their significance discussed. Two recent hypotheses are also considered: (1) that sapropels are absent in the western Ionian Sea; and (2) that stagnant phases were not basin-wide phenomena⁸. These latter points

contradict prevailing hypotheses^{4,5}. Information available in the Ionian Sea must be compared with data in adjacent basins to interpret properly late Pleistocene and Holocene physical oceanographic events in the Mediterranean.

The general spatial distribution of sapropels in the Ionian Sea can be determined from a survey of available cores and of published core data. The location of selected sapropel-bearing cores in the central and eastern Mediterranean (Fig. 1) indicates that Quaternary stagnation layers occur throughout the Ionian. These units are particularly well developed on the Mediterranean Ridge^{9,11}, and are also recovered in the Messina Basin plain (Pleistocene sapropels in JOIDES Leg 42A-Site 374 (ref. 12)) and in the western Hellenic Trench¹³. Evidence of stagnation phases to the north and east of the Ionian is recorded by the sapropels in adjacent deep basins of the Adriatic Sea¹⁴, Gulf of Corinth¹⁵, Aegean Sea¹² and the Levantine Basin^{1,6,7,16}.

Figure 1 also shows that cores displaying Quaternary sapropels occur in all external margins of the Ionian: in the westernmost sector, east of the Sicilian-Tunisian Platform and the Malta Escarpment (cores Lynch-II-3 (refs 17, 18) and Vema 14-136 (ref. 19)); in the northwestern sector, on the highly dissected Messina Cone south-east of the Strait of Messina and Calabria (cores R. D. Conrad 9-191 (ref. 1) and Vema 10-22, 68, 69 and 70 (ref. 19)); in the northern Ionian, in the Apulian and Strait of Otranto regions (University of Munich cores OT-25, 26 and 28 (ref. 20) and R. D. Conrad

9-190 (ref. 1)); in the southwestern Ionian (Atlantic Seal core 6-6 (ref. 21)); and in the southeastern Ionian (Chain core 61-31 (ref. 22) and Atlantic Seal core 6-5 (ref. 21)).

In contrast, cores collected on the Sicilian-Tunisian Platform, in the Strait of Sicily proper and in the various basins of the western Mediterranean do not display readily recognisable sapropels^{17,18}. However, there is some evidence, in the form of darker (olive-grey) layers containing pyritised burrows, of regionally important reducing conditions that affected these western basins^{23,24}. Radiocarbon dating of these layers shows that these partially anaerobic phases in the Tyrrhenian, Ligurian, Balearic and Alboran Seas may correlate with those that resulted in the formation of the eastern Mediterranean sapropels¹⁷.

Determination of regional facies distributions based primarily on detailed examination of lithofacies and ¹⁴C dating of core samples indicates that some of the major late Quaternary sapropels in the eastern Mediterranean extend from the Nile Cone in the Levantine Basin to the western Hellenic Trench-western Mediterranean Ridge sector^{5,13}. It is proposed here that the upper three sapropels identified in the Levantine Basin^{7,16} and in the eastern Ionian extend as far west as (but not on) the eastern margin of the Sicilian-Tunisian Platform. Radiocarbon dated samples of core Lynch-II-3 in the western Ionian identify these upper three sapropel layers (Fig. 2); they can be correlated with dated stagnation layers more than 1,000 km to the east (S₁, ~7,000–9,000 yr BP; S₂, ~23,000–25,000 yr BP; and S₃, ~38,000–40,000 yr BP (ref. 7)).

Sapropels in the Ionian and elsewhere are best displayed in those physiographical settings that favour the accumulation of enhanced proportions of suspensate and hemipelagic deposits relative to gravitative units. The well-defined sapropels in most cores recovered on the Mediterranean Ridge show that low accessibility to turbidites and mud flow deposits²⁵ favours well-defined sapropel layers. In contrast, sapropels are poorly developed (thick and indistinct) or not observed at all in some cores of the western Hellenic Trench¹³, on the Messina Cone and on the Libyan-Gulf of Sidra margin. This does not necessarily imply incomplete basin-wide stagnation phases as has been suggested by Thunell *et al.*⁸ A more likely explanation provided by detailed petrological examination (SEM, X-radiography) of such cores is that turbidites and other gravitates, indicative of high terrigenous input and sedimentation rates, locally have masked stratigraphic horizons that would normally display stagnation layers¹³. This 'dilution' phenomenon generally occurs in regions that have received a large sediment supply. The effect of gravitative incursions is observed in core Lynch-II-3 where sapropel S₂ is broken into three dark layers by silt and mud turbidites (Fig. 2). Locally, slumping has also resulted in removal of sapropel-bearing sections^{4,13}. Thus, sapropel and gravitate deposition are not mutually exclusive.

Water depth is another factor that must be considered in interpreting the spatial configuration of sapropels: the regional core survey in the Ionian indicates that stagnation layers form primarily at depths in excess of ~800–1,000 m where density stratification, oxygen content and reduced flow favoured anaerobic conditions on the sea floor^{14,17,20}.

Although the upper three late Quaternary sapropels (S₁, S₂, S₃) may be traced across most deep sectors of the eastern Mediterranean^{7,13,18}, we must note the presence of sapropels formed locally and realise that these can confuse regional correlation. Examples of apparently anomalous units resulting from local-restricted conditions and from slumping are identified in some sectors of the western Hellenic Trench¹³. Physical oceanographic parameters are significantly influenced by the physiographical complexity of this province which is broken into a series of deep (>4,000 m), isolated basins bounded by steep slopes and cobblestone topography²⁵. This highlights the need for working with a sufficiently dense core network and using sedimentological methods and an absolute age framework to correlate reliably over broad areas.

Fig. 1 Chart of the Ionian Sea showing the location of sapropel-bearing cores. Submarine features: STP, Sicilian-Tunisian Platform; ME, Malta Escarpment; MC, Messina Cone; MP, Messina Basin plain; MR, Mediterranean Ridge; SO, Strait of Otranto; GS, Gulf of Sidra; WHT, western Hellenic Trench. Surrounding land areas include: Libya and Cyrenaica (Cy), Crete (Cr), the Peloponnese (Pe), Greece (G), Sicily (S) and southern Italy (Calabria = Ca). Core symbols are: AS, RV Atlantic Seal (cruise 6-1967 (ref. 21)); C, RV Chain core 61 (1966 cruise, Woods Hole Oceanographic Institution^{16,22}); J, Joides (DSDP cruises 1970-Leg 13 and 1975-Leg 42A (refs 10, 12)); Ly, USNS Lynch (1972 cruise II, US Naval Oceanographic Office¹¹); M, RV Meteor (1969 and 1971 cruises, Deutsche Forschungsgemeinschaft⁹); Ma, RV Marsili (1976 cruise, Italian C. N. R.¹⁵); RC, RV Robert D. Conrad (1965-cruise 9, Lamont-Doherty Geological Observatory¹); T, Otranto Expedition (1968 cruise, University of Munich²⁰); Tr, RV Trident (1975-cruise 172, University of Rhode Island^{13,25}); V, RV Vema (cruises V10-1965 and V14-1958, Lamont-Doherty Geological Observatory^{15,19}).

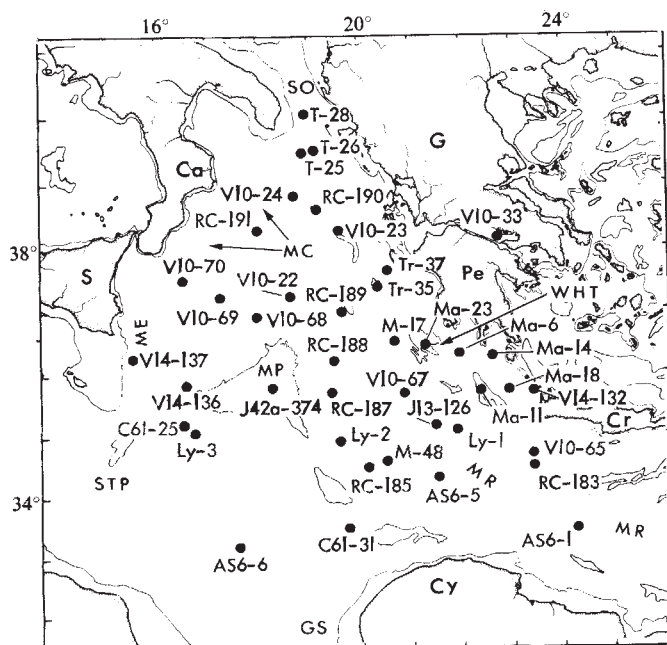
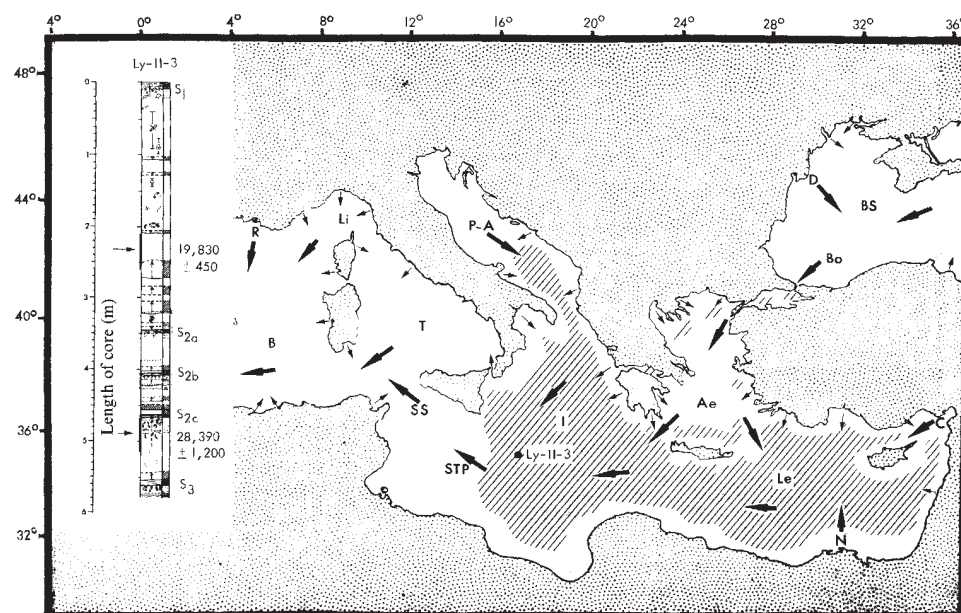


Fig. 2 Surface flow patterns in the Mediterranean Sea during late Quaternary stagnation phases. The hatched area shows the regional distribution of well-developed sapropels. Heavy arrows indicate surface water flow during periodic phases of excess less saline input, marked water mass stratification and sapropel formation. Also emphasised is the influence of fluvial input from large rivers such as the Danube (D), Nile (N), Ceyhan-Seyhan system (C), Po-Adriatic system (P-A) and Rhône (R) and smaller but more numerous fluvial systems (small arrows). The prevailing surface flow is from the Black Sea (BS) through the Bosphorus (Bo) and Aegean Sea (Ae), and then southwards into the Levantine (Le) and Ionian (I) seas. A predominant westwards trend is shown on the Sicilian-Tunisian Platform (STP) and in the Strait of Sicily (SS), Tyrrhenian (T) and Ligurian (L) seas and Balearic Basin (B). The log of core USNS Lynch-II-3 in the western Ionian Sea records the presence of the three most recent major sapropelic episodes (S₁, S₂, S₃); the arrows on the side of the log show the position of radiocarbon dated samples (dates in yr BP).



of core USNS Lynch-II-3 in the western Ionian Sea records the presence of the three most recent major sapropelic episodes (S₁, S₂, S₃); the arrows on the side of the log show the position of radiocarbon dated samples (dates in yr BP).

The large number of radiocarbon dated cores and available core density (Fig. 1) show that the upper three major sapropel layers in the eastern Mediterranean extend between the Nile Cone⁷ and western Ionian¹⁸; it remains to be shown that these units are not strictly isochronous east of the Strait of Sicily. Although the ages of these late Quaternary units are generally consistent throughout the area, it is possible that a more precise dating method will show that these sapropels vary in age regionally. A stagnation layer related to oceanographic conditions on the sea floor may reveal differences of tens to several hundreds of years or more between core sites separated by more than 1,000 km. Within the same region a sapropel layer could also record a different age between its position in a deep basin and on the adjacent shallow upper slope. Such an age difference would record the time of lateral migration of anaerobic conditions as well as vertical changes of the reducing level with time on the basin margins (as in the Black Sea²⁶).

The observed distribution patterns of late Quaternary sapropels favour the hypothesis of periodically altered basin-wide thermohaline circulation entrained by regionally important climatic and eustatic changes. Climatic fluctuations in the eastern Mediterranean considerably modified the precipitation-evaporation balance in an as yet ill-defined manner. At present, it is reasonable to relate the broad coverage of sapropels in the eastern Mediterranean (Fig. 2) to significantly increased input of less saline water, the formation of a well defined surface layer and marked density stratification of the water column. This less saline surface layer was almost certainly influenced by the change in precipitation as well as the Black Sea overflow suggested by Olausson⁴ and Ryan⁵, and the increased run-off from large rivers (such as the Danube, Nile, Ceyhan and Po) as well as from the numerous smaller fluvial systems (Adriatic, Turkish and others that together account for a significantly large total flow volume) draining the land-masses north of the eastern and central Mediterranean (Fig. 2).

The three upper radiocarbon dated sapropels in the western Ionian and adjacent regions provide evidence that the last stagnation phases were basin-wide events of relatively short duration (probably <2,000 yr). The absence of sapropels in the intermediate and deep basins on the Sicilian-Tunisian Platform

indicates that the sea floor of this elongate sill remained ventilated while anaerobic conditions prevailed below 1,000 m on the Malta Escarpment and in the Ionian Sea and regions to the east. Flow through the Strait of Sicily was not blocked, and the zone between Tunisia and Sicily continued to be the major passage for water masses moving between the eastern and western Mediterranean. The deposition of sapropel layers that are laterally continuous between deep (up to 5,000 m) basin floor and upper slope environments requires physical oceanographic conditions that do not conform to the 'minimum oxygen layer' model proposed for the origin of some sapropel deposits in other world oceans²⁷. More likely, the distribution patterns of late Quaternary sapropels in the Ionian and their extension to the east are directly related to episodes of what may be broadly recognised as an 'estuarine-type' circulation system. Density stratification was associated with possible short-term current reversals including the westwards flow of less saline surface water¹⁷. This palaeoceanographic scheme, previously outlined for the central and western regions of the Mediterranean^{23,28} is an appealing hypothesis that should be tested further.

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Contrasts between oceanic and continental 'unconformities' in the Oligocene of the Australian region

WIDESPREAD ocean floor unconformities around Australia have been explained by nondeposition of sediment and erosion by deep-sea currents, which undoubtedly occurred. The irregular duration of their hiatuses is in contrast to the widespread, brief but regularly terminated, early Oligocene unconformity on the continent. It is suggested here that the Palaeogene unconformities in Australia and the surrounding ocean basins have been derived from two different and possibly unrelated causes.

The concept of regional unconformities (*auct.*) in the deep ocean basins near Australia¹ has been developed^{2,3} with emphasis on their being the result of the scouring of the sea floor by initially northwards-flowing and later eastwards-flowing bottom currents, which came into existence in the Oligocene as a result of the cooling of Antarctic surface water and the initiation of thermohaline circulation. It has also been postulated that the onset of this cooling occurred close to the Eocene/Oligocene boundary and the first appearance of ice-rafted continental debris in high southern latitudes in the late Oligocene⁴ suggests the onset of glaciation at this time, but not of ice-cap formation⁵, which would be incompatible with floral evidence⁵. The Marshall Paraconformity⁶ has been postulated as a feature, not only of the southern ocean basins, but also of the southern continents, deep-sea erosion having been suggested as its cause.

In the deep ocean basins of the Australian region (Fig. 1), most well sections at Sites of the Deep Sea Drilling Project (DSDP)⁷ show a hiatus which includes the early Oligocene, but in many, the hiatus is of longer duration (Table 1). However, at many sites, late Eocene sediments occur below the hiatus, but the age of the beds above it shows no regional constancy. Where Palaeogene unconformities occur on the continent, the beds beneath the unconformity surfaces are variable in sedimentary facies but most are of late Eocene age and the hiatus is terminated by remarkably uniform and contemporaneous transgressive marine sediments which extend across much of the southern and southwestern part of the continent. Hiatuses in DSDP holes may be accentuated by local events, for instance many of the Indian Ocean holes recorded here were drilled on basement highs, even though some were within abyssal plain basins, where nonconformable overlap and perhaps unconformity has excluded parts of the sequence. No hiatus in a

DSDP hole should be accepted necessarily as representing the full duration of a regional unconformity, unless supported by evidence from seismic reflection profiles in the vicinity confirming the likelihood of the regional unconformity at that place. DSDP site 261 is a case in point, being sited on a basement high so that the oldest sediments and basalt could be reached with a minimum of drilling, it records a hiatus between Upper Cretaceous and Upper Miocene sediments. However, in the vicinity, seismic reflection profiles show considerably thicker Cainozoic sections and nonconformity against the basement high, of sedimentary units apparently equivalent to part of the hiatus. No site of DSDP legs 26 and 27 quoted (Table 1) was drilled where a complete or even substantial section of Cainozoic sediments was likely to be encountered. Kennett and Watkins⁸ have presented other evidence of sediment winnowing in the south-eastern Indian Ocean. The remarks presented here are not intended in any way to detract from the thesis of deep-sea erosion where the ocean basins are concerned, but rather to show that evidence on the Australian continent suggests different Oligocene events, attributable to causes other than erosion by deep-sea currents. The lack of correspondence, in time, of the hiatuses in the ocean basin sections, is also emphasised here.

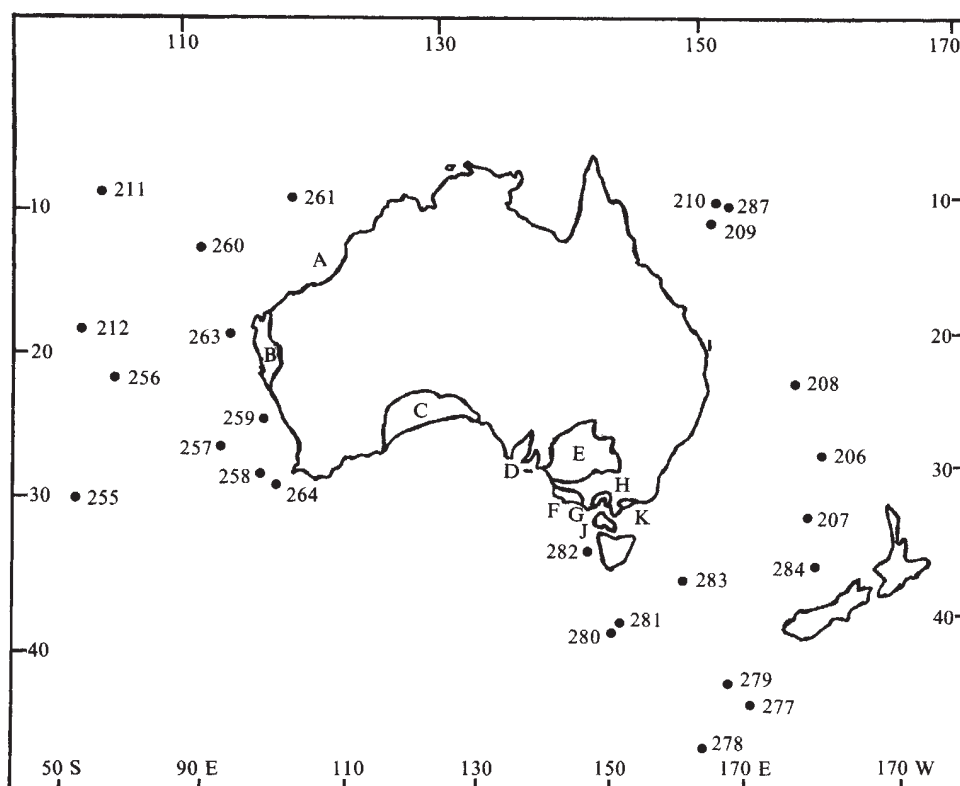
On the Australian continent, a paraconformity/disconformity occurs between Upper Eocene and Upper Oligocene rocks in the Carnarvon⁹ and Eucla¹⁰ Basins and on the north-west shelf⁹, the three westerly Tertiary basins of the Australian continental margin. Further east, in the St Vincent^{10,11}, Murray¹⁰, Otway¹², Aire¹³, Bass¹⁴, Port Phillip^{15,16} and Gippsland^{17–19} Basins, nonmarine sand or sandy clay, estuarine carbonaceous siltstone or coal measures underlie a pronounced diastem. In the Aire Basin, estuarine carbonaceous siltstone is underlain by marine beds containing *Globigerina angiporoides* Hornibrook, of very early Oligocene age, locally narrowing the estimated duration of the hiatus and supporting the dating of the onset of the regression given by Vail *et al.*²². There is no evidence that the sea entered the present-day on-shore parts of the Port Phillip and Gippsland Basins until the late Oligocene,

Table 1 Durations of Hiatuses at DSDP Sites adjacent to Australia, as cited in original core descriptions

Region	Leg	Site	Duration of Palaeogene hiatus
Indian Ocean	26	255	Middle Eocene–Lower Miocene
		256	? Upper Cretaceous–Pliocene
		257	Cretaceous–Quaternary
		258	Coniacian–? Upper Miocene
	27	259	Lower Eocene–Quaternary
		260	Middle Oligocene–late Middle Miocene
		261	Upper Cretaceous–Upper Miocene
		263	Lower Palaeocene–Upper Pliocene
	28	264	Upper Eocene–Upper Miocene
	22	211	Undated post Cretaceous–Pliocene
		212	Middle Eocene–Middle Miocene (unfossiliferous, deep-water zeolitic clay in interval)
Tasman Sea and Coral Sea	29	277	Late Oligocene–Early Pleistocene
		278	No Palaeogene unconformity
		279	All sediments post early Miocene
		280	Oligocene–late Miocene
		281	Late Eocene–early Miocene
		282	Mid Oligocene–early Miocene
		283	late Eocene–? Miocene
		284	All sediments post late Miocene
	21	207	Mid Eocene–early Miocene
		206	Late Eocene–Middle Oligocene
	30	208	Mid Mid Eocene–late Oligocene
		209	Late Eocene–? late Oligocene
		210	No unconformity
		287	Lower Oligocene–early Miocene

Uncored intervals are included. Sites are arranged with a roughly northerly trend.

Fig. 1 Australia, showing DSDP Sites adjacent to the continent, also the marginal Tertiary Basins: A North-west Shelf; B Carnarvon; C Eucla; D St. Vincent; E Murray; F Otway; G Aire; H Port Phillip; J Bass; K Gippsland.



where the unconformity below the late Oligocene transgressive sediments is as clear stratigraphically¹⁷ as elsewhere in Australia. The Gippsland Basin, flooded by the sea after the diastem but not before, seems to have subsided at approximately the time of the regression. This could be the cause of the reduced seismic expression of the unconformity in Gippsland and the interpretation of lessened regression given by Vail *et al.*²².

The paraconformity/disconformity is a major, continent-wide feature and in all the now-continental marine Tertiary Basins of Australia, the hiatus is terminated by contemporaneous late Oligocene marine transgressive sediments of calcareous clay or biogenic limestone facies, basally glauconitic in Gippsland and phosphatic in the Aire and Otway Basins. Their regularity is in great contrast to the temporally irregular termination of the diastems in the deep-sea sections. The change in pattern of major ocean currents postulated for the late Oligocene³ seems unlikely to foster deposition in the Australian shelf basins in the late Oligocene, after the former current regime had prevented it in the early Oligocene; particularly so as the younger, eastwards-flowing current system would seem more likely to erode the southern margin of Australia than previously. At DSDP site 282, the site closest to the southeastern Australian margin, the unconformity occurs contemporaneously with the transgression on the continent.

Some supplementary explanation such as a eustatic fall and subsequent rise of sea level, which has been invoked for localised instances^{9,19}, seems necessary to explain the whole continental situation. Evidence of it would occur on the continental shelves of the time and not necessarily in the deep ocean basins. Antarctic glaciation does not provide a cause for eustatic fluctuations of sea level at this time³⁻⁵. The production of the same effects by isostatic causes would involve regular movement, first up and then down, but without subsidiary fracturing, of 5,000 km of continental margin partly extended east-west and partly north-south, which is an unlikely expectation. Diastems, being sedimentologically negative features, do not receive prominence in the literature, but attention has been drawn⁶ to a diastem in the early Oligocene in all the

southern continents and New Zealand; there is a suggestion of a regressive phase at this time in the Tertiary (Lower Zamborian) of California²⁰ and the correlation charts of Eames *et al.*²¹ show the early Oligocene to be widely unrepresented by sedimentary formations in on-shore sections, in many parts of the world.

Vail *et al.* have deduced from geophysical data many regressions which they attribute to relative falls in sea level, particularly between the Jurassic and the present time. Their graphical quantification of these regressions shows the event in the early Oligocene to be the most intense of all those recorded. The Australian unconformity at this time, including that in Gippsland, is compatible with their conclusion²², but the ocean basins around Australia present a different history of deposition and nondeposition.

In the absence of glacially controlled eustasy at this time, the suggestion that transgressions and regressions are due to swellings and subsidences of the oceanic crust is generally plausible^{22,23}, but is discounted for the early Oligocene regression²² because of its short duration. The worldwide early Oligocene regressive event awaits fuller definition and explanation.

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Sunset and the orientation of a nocturnal migrant bird

THE information required for compass orientation among migratory birds is potentially available in the form of numerous environmental stimuli or cues¹. Most passerine migrants complete their annual migrations at night, and their orientation is thought to depend on a star compass or one based on geomagnetic stimuli¹. The pioneering work of Kramer² suggested that the setting sun may play a part in the migratory orientation of birds, but this possibility attracted little attention until more recently¹. I present here the first experimental evidence that directional information at the time of sunset plays a crucial part in the compass orientation of a nocturnal migrant *Passerculus sandwichensis*, the savannah sparrow.

The experiments were conducted near Clemson, South Carolina during the spring and autumn of 1977. Individuals mist-netted during the winter in the Clemson area were used in spring orientation tests, and experiments carried out in the autumn involved birds netted in North Dakota at the onset of their autumn migration. The birds were housed under natural photoperiod in an outdoor aviary, and tested only after they showed migratory restlessness (*Zugunruhe*) in activity cages and exhibited subcutaneous fat deposits³. (*Zugunruhe* is regarded as a measure of migratory readiness in caged birds⁴.) Orientation behaviour was examined by placing birds in circular, funnel-shaped cages and recording their locomotor activity using the Emlen 'footprint' technique⁵.

Controls were birds placed in the test cages at sunset as the Sun neared the horizon and were also exposed to stars (SS/ST situation). To determine to what degree access to sunset was necessary for appropriate migratory orientation the same individuals were tested under clear, starry skies without exposure to solar cues (ST situation). The birds were also tested under overcast night sky but with test exposure to sunset (SS/NO ST situation) to evaluate whether or not directional information at sunset was sufficient. All birds regardless of experimental situation were removed from their test cages and returned to the aviary approximately 2 h after the end of twilight (defined here as the period following the decline of the sun below the horizon and before the appearance of stars). Seven sparrows in the spring and seven in the autumn were tested at least once under all three situations and thus comprise my sample. All experiments conducted in the moonlight were excluded from this analysis, because of the possible biasing influence of moonlight on orientation.

A northerly directional preference is characteristic of spring orientation in this species and is evident in the orientation records examined in this study (Fig. 1 and Table 1). Six of seven sparrows exhibited statistically significant ($P < 0.05$) orientation in the spring when tested under the SS/ST situation and were orientated in a seasonally appropriate northwards direction, with the exception of one of three tests of S02 where the bird orientated to the east ($\bar{\alpha} = 83^\circ$). Although the orientation activity of S06 was generally northwards, the mean heading of only one of four SS/ST tests was statistically significant. When these seven individuals were tested without exposure to sunset

cues (ST situation) orientation activity decreased (Table 1) and lacked statistically significant directionality (Fig. 1). In fact, only S07 displayed a pattern of activity with a northerly bias. Two of the birds (S17 and S26) failed to show *Zugunruhe* according to my criterion. When S17 and S26 as well as S07 were tested a second time under the ST situation, this time in the presence of

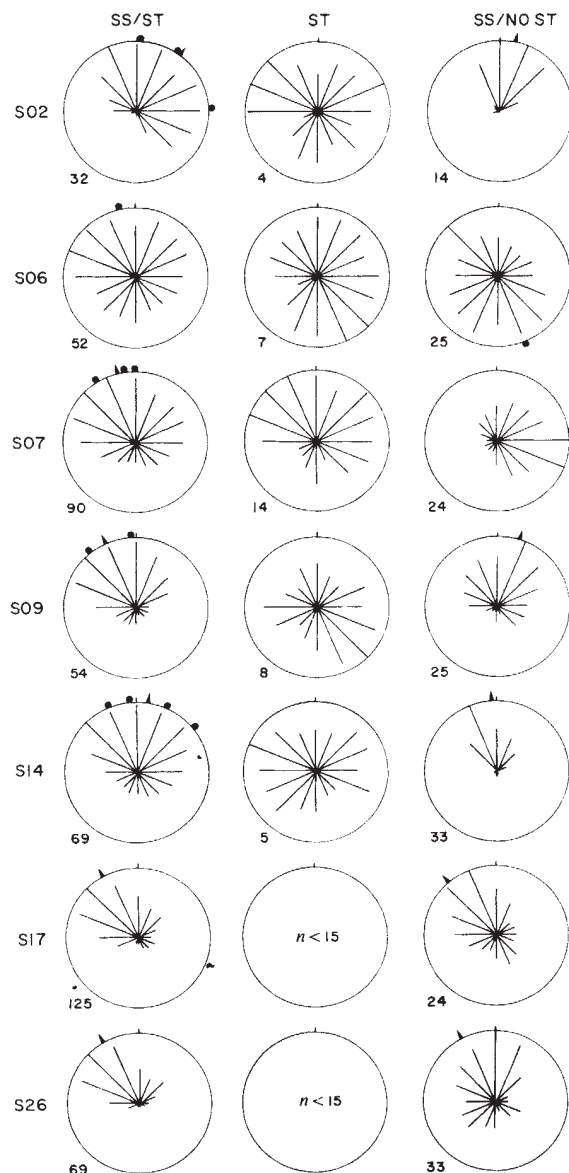


Fig. 1 Orientation behaviour of seven savannah sparrows tested in spring under varying exposure to sunset and stars. Vector diagrams are drawn such that the radius equals the greatest number of activity units in any one 22.5-degree sector. This number is given at the lower left of each diagram. The arrow on the circumference indicates the mean direction of orientation activity when the distribution is significant ($P < 0.05$). Replicate tests under a given experimental situation were combined for an individual and analysed accordingly. A dot on the circumference of a diagram represents the mean direction of an individual test if significant. Quantification of the orientation records closely followed Emlen and Emlen⁵. A scale of activity ranging from 1 to 30 was created from actual footprint records and used to numerically quantify activity. Mean direction ($\bar{\alpha}$) of activity for each bird under the various test situations was calculated by vector analysis⁶ and the circular distribution of activity statistically tested according to the Rayleigh test⁶. Because extremely low cage activity may not be related to migratory behaviour, a minimum level of nightly activity ($n < 15$) was set for analysis of a bird's orientation. In all diagrams north is 0° or 360° and is towards the top of the figure. See Table 1 for pertinent statistics.

the moon to the south, all activity was directed towards the moon azimuth (unpublished data). Comparing the orientation behaviour from these two test situations (SS/ST and ST) affirms the significance of the sun as a cue and indicates that it is necessary for appropriate migratory orientation in this species.

Examination of the SS/NO ST orientation diagrams (Fig. 1) for five of the seven spring birds suggests that cues at the time of sunset are sufficient for appropriate orientation in the experimental conditions. Only two birds failed to show either seasonally suitable directionality (S07) or activity concentrated enough to be deemed significant (S06). Otherwise the orientation corresponded with the direction assumed when both sunset and stars were present.

The results of the autumn experiments are equally convincing. All seven savannah sparrows exhibited well orientated behaviour in seasonally meaningful directions (Fig. 2 and Table 2); six displayed southerly preferences while F33 headed in a southwesterly direction ($\bar{a} = 237^\circ$). When these birds were denied sunset cues at the time of testing but allowed access to stars (ST situation), weak ($P > 0.05$) or seasonally inappropriate orientation behaviour was typical. In two instances, however, southerly preferences were recorded under stars alone: F13 ($\bar{a} = 180^\circ$) and one of two ST tests of savannah F23 ($\bar{a} = 186^\circ$). In the latter bird the combined results produced no significant orientation. These results do suggest that at least one and possibly two of the seven sparrows tested in the autumn may be capable of migratory orientation in the absence of solar information. However, similar unpublished data from other experiments argue strongly against this possibility.

The directional activity in the fall under the SS/NO ST situation corresponded for the most part to that found in the control (SS/ST) situation (Fig. 2). Two exceptions were noted; F33 was not well orientated although the activity pattern suggests a southwesterly trend comparable to that shown under SS/ST, and F13 was inactive. In autumn as in spring, most sparrows were most active when exposed to both sunset and stars (Table 2).

Table 1 Orientation behaviour of savannah sparrows in the spring under varying exposure to sunset and star cues

Bird	Test	<i>n</i>	$\bar{a}$ (deg)	<i>r</i>
S02	SS/ST	109(3)	37	0.406*
	ST	17	—	0.071
	SS/NO ST	21	13	0.846*
S06	SS/ST	230(4)	330	0.114
	ST	32(2)	97	0.109
	SS/NO ST	102(2)	205	0.109
S07	SS/ST	351(4)	351	0.236*
	ST	60	—	0.207
	SS/NO ST	70	83	0.356*
S09	SS/ST	165(2)	336	0.412*
	ST	30(2)	—	0.116
	SS/NO ST	68	17	0.421*
S14	SS/ST	257(4)	9	0.276*
	ST	21	—	0.084
	SS/NO ST	45(2)	356	0.649*
S17	SS/ST	302(5)	330	0.447*
	ST	2	—	—
	SS/NO ST	69	319	0.259*
S26	SS/ST	32	330	0.637*
	ST	6	—	—
	SS/NO ST	30	332	0.263*

Sample size (*n*) for a given test was determined by dividing the total number of units of footprint activity by 8 for independence of the data measures⁷. When a bird was tested more than once under a given situation it was noted parenthetically and the total *n* was divided by the number of tests to acquire an average *n* for a situation. $\bar{a}$, Mean direction of orientation activity. *r*, Rayleigh's measure of concentration.

* Statistically significant at the 95% level.

Circumstantial evidence already suggests that solar cues may be important and consequently supports my experimental results. For the sake of brevity, I only outline some of this evidence. (1) The existence of a sun compass has been documented in several night migrants^{7,8}. (2) The peak of migratory activity occurs at night among nocturnal migrants, but the actual night's migration usually commences before the end of twilight^{9,10} and probably on many occasions before any stellar cues are available. (3) Passerine nocturnal migrants are known to make sea crossings¹¹ which necessitate flying beyond darkness during daylight hours¹⁰. (4) Radar observations¹² indicate that artificially released white-throated sparrows (*Zonotrichia albicollis*) assume appropriate migratory directions during twilight before stars are visible. (5) Studies of migrants in field conditions show well orientated behaviour despite overcast

Fig. 2 Orientation behaviour of seven savannah sparrows tested in autumn in varying exposure to sunset and stars. The overcast for the SS/NO ST situation in the autumn was artificial; circular boards were placed over the tops of the cages. The boards were positioned in such a way that the bird could see the setting Sun and twilight but was unable to view the stars. See Fig. 1 for details and Table 2 for statistics.

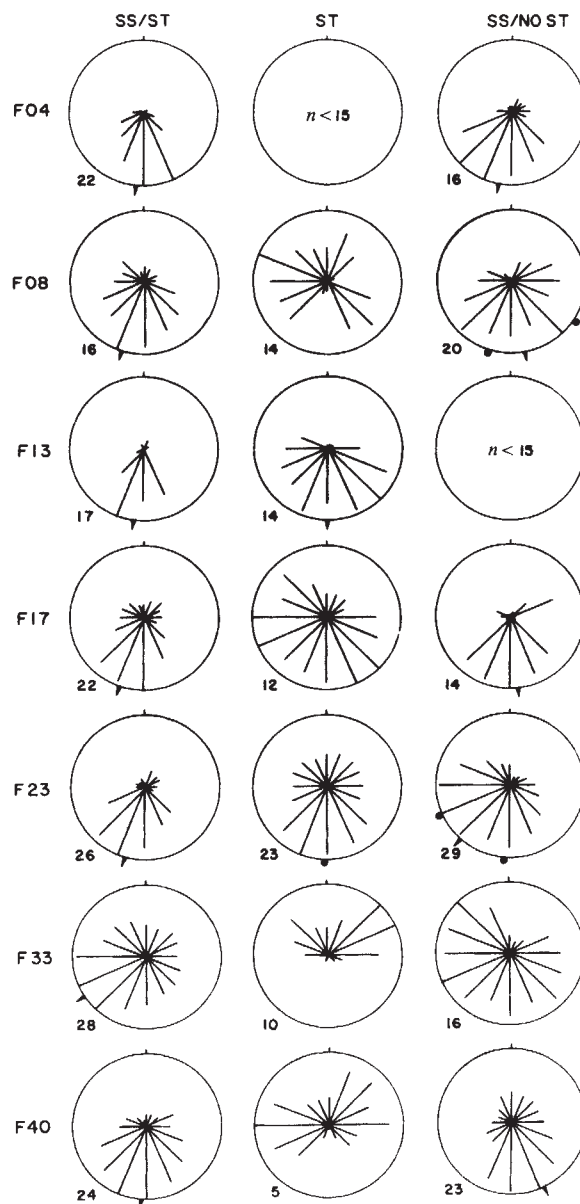


Table 2 Orientation behaviour of savannah sparrows in the fall under varying exposure to sunset and star cues

Bird	Test	n	$\bar{a}$ (deg)	r
F04	SS/ST	37	186	0.781*
	ST	2	—	—
F08	SS/NO ST	39	189	0.541*
	SS/ST	42	198	0.433*
	ST	46	—	0.157
F13	SS/NO ST	63(2)	170	0.363*
	SS/ST	22	189	0.739*
	ST	40	180	0.523*
F17	SS/NO ST	2	—	—
	SS/ST	56	199	0.378*
	ST	49	—	0.220
F23	SS/NO ST	32	174	0.514*
	SS/ST	59	197	0.466*
	ST	84(2)	—	0.189
F33	SS/NO ST	98(2)	222	0.368*
	SS/ST	104	237	0.213*
	ST	22	26	0.520*
F40	SS/NO ST	68	—	0.196
	SS/ST	68	186	0.445*
	ST	16	—	0.167
	SS/NO ST	70	154	0.293*

See Table 1 for more details.

* Statistically significant at the 95% level.

night skies¹³⁻¹⁶. (6) Other studies^{13,17,18} have pointed out that migrants displayed appropriate orientation under overcast skies if the Sun had been visible before the nightly departure. (7) Migrants have also been observed to exhibit orientated migratory behaviour in the morning hours, possibly as a corrective measure for displacements during a night's flight¹⁹. (8) Even when stellar information is available, migrants may depend on input from other cue systems for their initial selection of a migratory heading, for example, the Wiltschkos²⁰⁻²² present experimental evidence that the star compass of the European robin (*Eriothacus rubecula*) is dependent on its magnetic compass. Finally, additional support comes from recent autumn orientation experiments with white-throated sparrows, another North American nocturnal migrant (V. P. Bingman and K. P. Able, personal communication). Significant directionality was seen only after the white-throats were exposed to the setting Sun along with stars. Such results, like those reported here, are reminiscent of Kramer's early work² where he found that his caged nocturnal migrants assumed proper directions at night only after seeing the Sun at or before sunset. That others¹ have reported well orientated behaviour under stars alone for other species must in part be attributed to species-specific orientation systems.

Evidence^{1,23} in the field of avian migratory orientation suggests the existence of multi-cue systems of orientation. Such systems could assume at least two forms: first a redundant or cue-equivalent system wherein orientational information is dependent on the availability of cues and when possible pooling of information from a variety of sources takes place, or second, a hierarchical or weighted system wherein the bird relies on a primary orientation system from which other cue systems are dependent or calibrated. If solar and stellar cues are equivalent sources of compass information, migratory activity should be well orientated under either cue system. Such is not the case. Savannah sparrows orientate very poorly if at all in the absence of solar information at sunset, suggesting that this species does not possess an independent star compass. When the only visual cue available was sunset, most of the birds displayed orientation which agreed rather well with that recorded when both systems were present. Nonetheless, activity tended to be directionally more concentrated (higher *r* values) as well as more frequent in the appropriate direction (Figs 1, 2, Tables 1, 2) in the SS/ST situation. This implicates a

hierarchical arrangement between a primary cue (solar) and a secondary cue (stars). Quite possibly directional information is transferred from one cue system to another. Regardless of the exact nature of the solar-stellar relationship, I conclude that directional information at the time of sunset is both necessary and sufficient for migratory orientation in the savannah sparrow.

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Evoked potential evidence of adaptation to spatial Fourier components in human vision

INVESTIGATIONS of the nature of human spatial vision have yielded considerable evidence consistent with the hypothesis that the human visual system contains channels selectively sensitive to narrow ranges of spatial frequencies¹. Unfortunately, much of this evidence, which comes from studies of the detection of gratings, can also be explained in terms of the detection of single edges or lines². However, two studies^{3,4} of orientation-contingent colour aftereffects have involved attempts to discriminate between these explanations. Through the use of checkerboard stimuli, which have major Fourier components at 45° to the check edges⁵, these studies showed that chromatic adaptation occurs at orientations determined by the Fourier components of the adapting pattern and not by the edges present. After subjects had adapted to upright red and oblique green checkerboards, achromatic test gratings appeared pink when vertical or horizontal and green when oblique. Comparable aftereffects were observed when achromatic checkerboards were viewed after adaptation to

coloured gratings³. The strongest aftereffects were obtained⁴ for test gratings with a fundamental spatial frequency a factor of 1.5 above the periodicity of the adapting checkerboard; that is, close to the fundamental Fourier component of the checkerboard. We present here evidence, obtained from evoked potentials, that attenuation of the activity of orientation-specific cells in one region of the human visual cortex occurs largely in cells which are tuned to the orientations of the fundamental Fourier components of an adapting pattern rather than to the orientations of the edges contained in the pattern.

The transient visual evoked potential (VEP) elicited by the onset of a briefly presented pattern and recorded using electrodes attached to the scalp over the occipital lobe, has three major components occurring within 200 ms after stimulus onset⁶. We are concerned here with the initial component (CI, latency 75 ms) which Jeffreys^{6,7} has suggested originates in striate cortex, and which is sensitive to the orientation and size of the pattern⁸. We used an adaptation paradigm analogous to that used before^{3,4} but with achromatic adaptation patterns and much briefer presentation times. The experimental procedure has been described elsewhere⁸. The stimuli were high contrast photographic transparencies presented in a tachistoscope. Each stimulus cycle contained a 300-ms presentation of an adapting pattern followed, after an inter-stimulus interval which varied randomly between 100 and 200 ms, by a 25-ms presentation of the test pattern. During the inter-stimulus interval, and for 1 s after each presentation of the test pattern, an isoluminant uniform field was presented. The subject fixated a small, continuously present, central fixation cross. Each field subtended 9° in diameter and had a mean luminance of 64 cd m⁻².

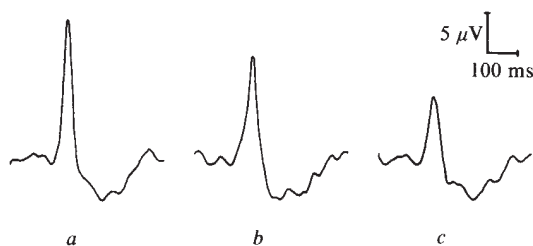


Fig. 1 Typical transient VEP waveform elicited: *a*, by a horizontal, 2.0 cycles per degree, grating in the left half of the field without adaptation; *b*, after adaptation to a horizontally orientated checkerboard; *c*, after adaptation to an obliquely orientated checkerboard. Subject V.W.

The VEPs elicited by the onset of the test pattern were recorded bipolarly between two electrodes attached to the scalp 5 cm on each side of the midline, 4 cm above the inion. The use of these electrode positions in conjunction with a test pattern confined to one vertical half of the field minimised modification of the amplitude of CI resulting from temporal overlap with the component which followed (CII)⁷. Responses to 80 successive presentations of the test pattern were averaged over 500 ms, commencing 100 ms before the onset of the test pattern. An analogue plot of amplitude against latency (Fig. 1) facilitated visual inspection of amplitude variations; in addition, for each plot a digital value of the mean amplitude during the 100 ms immediately before the onset of the stimulus (baseline amplitude) was obtained and subtracted from the peak amplitude occurring 70–80 ms after onset to give a measure of the amplitude of CI. This measure of CI was then expressed for each adapting condition as a percentage of the amplitude elicited during a control run in which the adapting pattern was replaced by a uniform field.

In the first experiment the test pattern was a horizontally orientated Esquare-wave grating of spatial frequency 2.0 cycles per degree. The adapting pattern was either a checkerboard with vertical and horizontal check edges and a check width equal to the bar width of the grating (15 min of arc), or an obliquely orientated checkerboard of an increased check size (21 min of arc) such that the diagonal of each check subtended

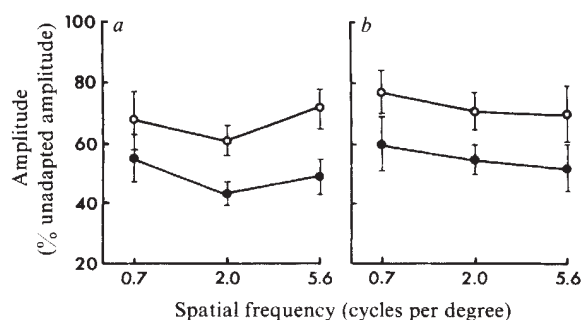


Fig. 2 Mean CI amplitude ($n=6$) elicited by: *a*, a horizontal grating of the size shown after adaptation to a horizontally orientated checkerboard (○) or an obliquely orientated checkerboard (●) of appropriate size; *b*, a checkerboard of the size shown after adaptation to a horizontal grating (○) or an oblique grating (●) of appropriate size.

30 min of arc. The fundamental Fourier components of the latter checkerboard lie at 2.0 cycles per degree in the vertical and horizontal directions. The VEPs elicited in these two conditions and by the same pattern without adaptation are illustrated for one subject in Fig. 1. Both adapting patterns cause amplitude attenuation but the effect is much more marked in the case of the oblique checkerboard.

The experiment was repeated using test gratings of spatial frequency 0.7 and 5.6 cycles per degree, the adapting check sizes being adjusted accordingly. Figure 2*a* shows the mean CI amplitude for six subjects for both adapting conditions at all three spatial frequencies. In each case more attenuation occurs after adaptation to the oblique than to the horizontal checkerboard.

In a similar experiment the test pattern was a checkerboard with vertical and horizontal sides to each check. The adapting pattern in this case was a square-wave grating, either horizontally orientated and with the same bar width as the test pattern check width, or obliquely orientated and with the spatial frequency of the fundamental Fourier component of the test checkerboard, that is a factor of 1.4 above the spatial frequency of the horizontal adapting grating. Again this procedure was repeated with three different test sizes and corresponding adapting sizes. Figure 2*b* shows the mean CI amplitude for the six checkerboard test conditions. Both horizontal and oblique adaptation cause amplitude attenuation but significantly greater attenuation occurs in the case of oblique adaptation.

These results show clearly that at the level of the source of CI (thought to be in striate cortex) greater adaptation occurs at the orientations of the major Fourier components present in an adapting pattern than at those of the edges contained in the pattern. They therefore provide physiological evidence that visual stimuli are represented in the visual system at least partly in terms of their Fourier spectra. They do not, however, necessarily constitute evidence that spatial vision derives primarily from a Fourier transform of visual space. Psychophysical data exist⁹ that are difficult to explain in terms of narrowly tuned spatial frequency channels, and indeed certain properties of pattern VEP components originating in other regions of visual cortex are inconsistent with such a proposition¹⁰. Therefore it may be appropriate to regard spatial frequency content as one feature of the visual stimulus, and spatial frequency channels as one of several parallel feature-detecting mechanisms in the visual system.

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Perceptual effect of pursuit eye movements in the absence of a target

THE traditional explanation¹ of visual tracking has been that smooth tracking by the eyes depends on the presence of a moving retinal image at or near the fovea. However, evidence is steadily accumulating suggesting that this explanation is incomplete². The presence of an extrafoveal afterimage or stabilised retinal image in certain circumstances suffice to initiate smooth tracking movements³⁻⁵; observers are able to track, in the dark, the position of their own hand⁶ or of a moving sound source⁷ and may, after considerable practice, be able to generate sinusoidal tracking movements purely voluntarily^{4,8}. We describe here a perceptual effect apparently created by the generation of smooth horizontal tracking eye movements, in the absence of a target, by observers regarding a display of dynamic visual noise. We interpret this effect in terms of the presence within random visual noise of features which lead to the neuronal representation of movement at some unspecified level of organisation of the visual system.

We took part in the following experiment together with two inexperienced observers. The observer's head was restrained in the vertical position by a dental bite bar and by steel bars abutting against the forehead and temples. The observer was confronted by the image, reflected in a mirror placed at 45° to his line of sight, of a television monitor screen which subtended 14° of visual angle. Dynamic noise ('snow') was generated on the screen by connecting the monitor to a videorecorder which was switched on but not running. The observer's horizontal eye movements were recorded photoelectrically, by locating the temporal boundary of the pupil-iris-sclera (the limbus) in an infrared beam which was reflected by way of a fibre-optic guide on to a photodiode whose output was displayed on an oscilloscope screen⁹. The position of the incident beam, which was reflected to a greater or lesser extent as more or less of the pigmented iris intruded into the illuminated area, was adjusted with a micromanipulator to obtain a signal that varied linearly with eye position throughout the 14° subtended by the display. At intervals the monitor screen was placed on its side, so that the raster ran vertically, or returned to the horizontal position, a procedure which did not influence the results described below.

Each observer was instructed to attempt to generate smooth tracking eye movements in a horizontal direction to and fro across the display. Naive observers were aided in the early stages by moving a pencil tip across the screen and pointing out to them that a small area of noise near the tip appeared to move with it; they were then instructed to track this area of noise with the pencil removed. In contrast to the results described by Kommerell and Täumer⁴, who stress the considerable training required to generate sinusoidal tracking movements while regarding a static featureless display, we all learnt to produce smooth tracking after at the most one or two 15-min sessions (see Fig. 1).

It was a curious sensation to be tracking without being sure of what it was that one was tracking. The initial impression was of a vaguely defined area of the noise field, described by some observers as a vertical band, by others as a more circular shape, which perceptually detached itself from the background and moved back and forth across the display with the eyes. As soon as the observer ceased tracking, this 'target' disappeared. After

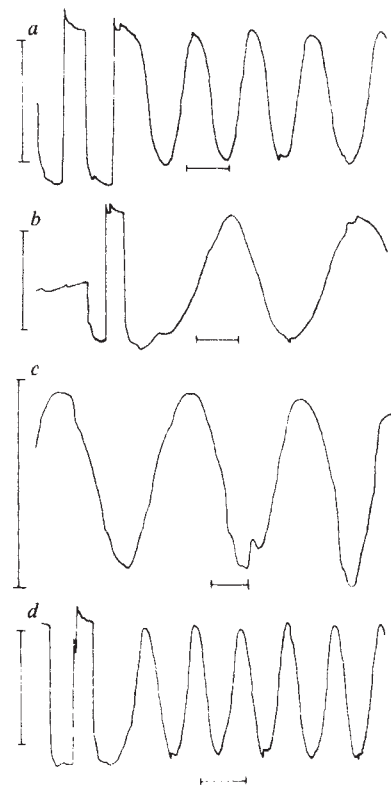


Fig. 1 Records of eye movements made while the observer was inspecting a field of dynamic visual noise. Horizontal markers represent 1 s and vertical markers represent 10° of visual angle. *a*, The display is not filtered in any way. The vertical lines at the start of the record are caused by saccades across the full width of the display, which the observer made to calibrate the system and to illustrate the difference between voluntary saccades and the smooth tracking which follows. *b*, Voluntary slow tracking by MJM of an unfiltered display. *c*, Tracking by RW of a low-pass filtered version of the display. *d*, Tracking by MJM with a neutral-density filter in front of one eye.

considerable practice we could maintain tracking with very little conscious impression of target motion at all.

The velocity of tracking movements maintained in this way was variable and could to a certain extent be consciously controlled. The initial tracking produced by observer RLM took the form of slow leftward pursuit at 3.3 deg s⁻¹, followed by a rapid saccade back to the right-hand edge of the screen; later in the experiment RLM showed smooth tracking in both directions at a velocity of 5.6 deg s⁻¹. Observer DS initially showed smooth pursuit at 5.5 deg s⁻¹ from left to right. Observer MJM could voluntarily control tracking speed at velocities between 8 and 27 deg s⁻¹; RW showed a fairly constant pattern of tracking in both directions at velocities between 6 and 10 deg s⁻¹. Smooth tracking could also be maintained when the high spatial frequencies of the noise were filtered out by placing a sheet of translucent paper in front of the screen (Fig. 1c).

The fact that our observers were able to generate tracking eye movements when inspecting the noise display may be interpreted in one of two ways. Either the effect is no more than another example of the ability of observers to generate, volitionally, smooth tracking eye movements^{4,8}, which seems unlikely in view of the small amount of practice required by our observers, or the generation of tracking is facilitated if not uniquely determined by some unspecified feature of the display. The following evidence suggests that such features may lead to the neuronal representation of movement at some level of organisation of the visual system.

Tyler¹⁰ described an illusion that is seen by an observer who looks at dynamic visual noise with a light-attenuating filter in front of one eye. If the filter is in front of the left eye, both eyes being open, the random movement of the dots on the screen is

replaced by coherent motion in which some dots appear to stream from left to right in a depth plane behind the screen, while other dots stream from right to left in a depth plane in front of the screen. If the filter is placed in front of the right eye, the mirror image of this impression is obtained. This is very possibly related to the result described earlier by Ross¹¹, in which computer-generated dots were presented to one eye slightly earlier than to the other; and at first sight both results resemble the well-known Fertsch-Pulfrich stereophenomenon^{12,13}, in which a target oscillating horizontally against a stationary background appears to move in a clockwise (as if viewed from above) orbit when inspected binocularly with a light-attenuating filter in front of the left eye, and in an anticlockwise orbit when the filter is placed in front of the right eye. The traditional explanation, due to Fertsch, is that the filter introduces a latency lag in the covered eye, so that a disparity between the two eyes is induced by motion of the target. As Tyler points out, the difficulty in applying Fertsch's explanation to the random-movement stereophenomenon is that there is no obvious stimulus to horizontal motion in the display. The reasons why Tyler's own explanation of the phenomenon he describes has to be rejected are complex and are fully explained elsewhere¹³; briefly, Tyler supposes that the filter introduces a delay such that dots seen in frame n by the unfiltered eye are paired with dots seen in frame $n-1$ by the filtered eye. Morgan and Thompson¹³ point out that this 'shifted pairing' explanation is plausible only if the filter introduces a delay of at least one half of the interframe interval (about 25 ms in the case of successive frames of a television picture): and yet the effect may be seen, as Tyler himself points out, with filters introducing delays no greater than 5 ms. These difficulties may be overcome if we suppose, in agreement with Mezrich and Rose¹⁴ and with Ross¹¹, that the random-noise stereophenomenon should be treated as a special case of the Fertsch-Pulfrich stereophenomenon in which the feature of horizontal motion is not consciously perceived until the eyes are differentially filtered.

Thus it is not surprising that our observers were able to sustain tracking when the noise display was viewed with a light-attenuating filter in front of one eye, since coherent motion was observed as described by Tyler. Either the apparently nearer or apparently further depth plane of drifting dots could be tracked. There was a tendency for the dots to appear to move more rapidly when tracked in this way than when viewed with the gaze held stationary and pursuit thus tended to be rather fast (Fig. 1d). The fact that the dots appeared to accelerate when pursued is very probably related to what Robinson³ describes as the 'Tantalus-like' pursuit of a stabilised extrafoveal retinal image as attempts are made to fixate it.

The perceptual segregation of a part of the display, contingent on the generation of tracking eye movements, shows considerable similarity to the observations reported by Lamontagne¹⁵ and by Heywood¹⁶: an observer who regards a stroboscopically illuminated row of dots is able, if the stroboscope flashes at the right frequency, to track along the row of dots, and obtains the impression of a single dot in motion. In both situations movement of the eyes fails to induce movement of contours on the retina, thereby producing the same feedback as that obtained when the eyes track a real moving target. The illusory effect begins to become comprehensible when we consider that smooth tracking may be controlled by mesencephalic rather than cortical mechanisms¹⁷, as it has been argued on other grounds¹⁸ that the visual mechanisms of the midbrain are more concerned with attributes such as direction and motion than with precise details such as shape.

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Light-regulated body ion balance in marine slug *Elysia viridis* (Montagu)

THE small, leaf-shaped marine slug *Elysia viridis* (Ophisto-branchia, Sacoglossa) owes its green colour to borrowed pigments. Unusual, although not unique in the animal kingdom, it is part of a 'chloroplast symbiosis' and is photosynthetically active. The chloroplasts, which are found in the cells of the highly branched digestive channel (hepatic diverticula), have been identified as originating from the siphonous green alga *Codium fragile*, on which *Elysia* feeds¹. Removed from its food source but kept in the light, *Elysia* can live for months by means of photosynthesis. In the dark, however, even when offered food, it soon dies². As part of the relationship the animal cells affect the morphological integrity and function of the symbiotic chloroplasts, and I report here that the cells themselves are influenced by the presence of the chloroplasts. The ion balance of *Elysia* is regulated by light through the mediation of the chloroplasts.

In several marine algae (including *Chaetomorpha linum*, *Nitella translucens*, *Valonia ventricosa* and *Hydrodictyon africanum*) ion fluxes (Na^+ , K^+ and Cl^-) are regulated by photosynthetically active light, probably through photophosphorylation³⁻⁶. Uncoupling of ATP synthesis by carbonyl cyanide *m*-chlorophenylhydrazine (CCCP) in illuminated cells of *Hydrodictyon africanum* strongly inhibits cation fluxes⁵.

Sodium and potassium ion transport was studied in live slugs by measuring fluxes of radioactive tracers (^{22}Na , ^{24}Na , ^{42}K and ^{86}Rb) under several intensities of white light and in darkness. Uptake took place from labelled artificial seawater simulating the slugs' natural medium (25‰ salinity, 338 mmol Na^+ , 7.2 mmol K^+). Slugs were collected in the Danish Limfjord and kept in aerated seawater aquaria together with *C. fragile* before use. Radioactivity was measured on a NaI crystal counter connected to a multichannel analyser.

Light enhanced both influxes and effluxes of Na^+ and K^+ , resulting in a more rapid turnover of ions in light than in darkness (Tables 1 and 2). However, the most significant stimulation was that of ^{24}Na efflux and ^{86}Rb influx (Table 2), the latter especially at higher intensities of light. The equilibrium concentrations that were reached after approximately 20 h of uptake did not differ significantly in light and darkness. The total body Na^+ (239 ± 50 mmol, in light; 264 ± 50 mmol, in darkness) and K^+ (26.7 ± 5 mmol, in light; 26.0 ± 5 mmol, in darkness) (averages based on four slugs in each group) corresponded to 0.75 and 3.7 times the respective concentrations in the surrounding seawater. This suggests active accumulation of K^+ and extrusion of Na^+ .

Table 1 Ion fluxes in *Elysia viridis*

Light	Darkness	Light	Darkness
²⁴ Na influx		²⁴ Na efflux	
11.7	10.4	140	52.6
28.2	23.4	187	67.3
46.9		237	76.6
		266	90.5
		²² Na efflux	
		175	137
		201	138
		229	164
		236	201
⁴² K influx		⁴² K efflux	
1.70	0.80	1.25	0.64
1.70	1.32	1.40	0.89
1.70		2.40	1.70
		3.90	2.20

Values refer to single animals. Ion fluxes are expressed as $\text{nmol s}^{-1} \text{g}^{-1}$. Light exposure was at $1.0 \text{ J m}^{-2} \text{s}^{-1}$. Statistical errors of the radioactive counting are $<1\%$. The temperature was kept constant by circulating water at $15 \pm 1^\circ \text{C}$ (^{24}Na and ^{42}K) and at $24 \pm 1^\circ \text{C}$ (^{22}Na).

The ^{86}Rb fluxes (equivalent to potassium) were studied as a function of the chlorophyll content (Chl *a* + Chl *b* determined in 90% acetone) of the animal. For each light intensity the rate of influx of Rb^+ increased with increasing chlorophyll concentration. In slugs with a very low chlorophyll content this influx rate was as low as in the dark controls (Table 2). Light-enhanced K^+ efflux, however, was not proportional to chlorophyll concentration.

Exposure to light or darkness for 1 h immediately before the uptake of radioactivity (^{86}Rb) also influenced the rate of uptake. Again light had an enhancing effect (results not shown), suggesting the utilization of a light-created energy reserve, probably in the form of ATP, for subsequent ion uptake.

In spite of the intense green colour of the slugs, the chloroplasts constitute a small fraction of total body volume (1–3% calculated from chlorophyll determinations). Thus the chloroplasts cannot be considered one of the slug's principle ion pools. Photoregulation of ion fluxes seems to occur across the animal cell membrane although the energy thus utilised is supplied in part by the photosynthesis of its symbiotic chloroplasts.

Table 2 Ion fluxes and chlorophyll content in *Elysia viridis*

Light ($3.0 \text{ J m}^{-2} \text{s}^{-1}$)	Light ($1.9 \text{ J m}^{-2} \text{s}^{-1}$)	Darkness
⁸⁶ Rb influx		
(0.075) 1.70	(0.197) 1.33	(0.411) 1.05
(0.277) 4.26	3.42	1.13
4.60	(0.274) 3.73	2.48
4.60	5.40	(0.560) 2.65
(0.410) 6.66		2.70
(0.430) 5.40		
⁸⁶ Rb efflux		
0.94		1.02
1.10		1.20
(0.274) 1.20		(0.560) 1.33
1.30		1.43
(0.075) 1.45		
(0.270) 1.54		
(0.197) 2.10		
2.55		

Values refer to single animals. Ion fluxes are expressed as $\text{nmol s}^{-1} \text{g}^{-1}$ and chlorophyll content (in parentheses) as $\mu\text{g mg}^{-1}$. The statistical errors of the radioactive counting are $<1\%$. Temperature was kept constant by circulating water at $24 \pm 1^\circ \text{C}$. The influx is proportional to the chlorophyll content whereas the efflux is not.

In plants, light-mediated electron, proton and ion transport occur across the chloroplast membrane⁷. Electron flow in photosynthesis causes an inward (into the chloroplast) flow of protons accompanied by a movement of cations in the opposite direction, which neutralises the charge movement of the protons taken up. Thus photosynthesis does not simply create a cation sink in the cytoplasm. The light-mediated K^+ influx and Na^+ efflux found in many plant cells need to be explained by another mechanism, possibly a more direct effect on the cytoplasmic membrane. It is not clear how communication is effected between the chloroplast and ion pumps at the plasmalemma⁷, and the same might apply to *Elysia*. In *E. viridis* light seems to have two effects: (1) the enhancement of ion fluxes in both directions, resulting in a more rapid turnover and, (2) more specific stimulation of Na^+ efflux and K^+ influx. The first might be due to an increase in membrane permeability. The second, which seems to be more directly linked to photosynthesis, as suggested by the enhancing effects of light intensity and chlorophyll concentration, might be due to stimulation of active ion pumping by ATP from photophosphorylation, although other mechanisms are possible.

The pigments extracted from *E. viridis* have similar chromatographic pattern and absorption bands to those of the photosynthetic pigments found in *C. fragile*⁸ (β -carotene, zeaxanthin, neoxanthin, violaxanthin, siphonoxanthin, siphonin, chlorophyll *a'* and chlorophyll *b*). This does not exclude the possibility that trace amounts of other pigments, even of animal origin (porphyrins are found in *Aplysia punctata* integuments⁹) participate as photoreceptors in the photoregulation of ion transport.

In conclusion, however, the findings reported here represent the first demonstration of the regulation of the ion balance of an animal by sunlight.

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Conservation of autosomal gene synteny groups in mouse and man

WHILE genes on the X chromosome have been conserved during evolution¹ little is known about the degree of conservation of autosomal synteny groups for species distantly related in evolution such as mouse and man. The mouse gene map based on sexual genetics^{2,3} has been expanded by analysis of interspecific somatic cell hybrids segregating mouse chromosomes, so that the genetic maps of man and mouse can be compared. The available information indicates that genes located on different arms of the same human chromosome are not syntenic in the mouse, and genes which are many map units apart (25–45 cM) in the mouse are unlikely to be syntenic in man^{4,5}. In contrast, genes that are tightly linked (less than 1 cM apart) seem to remain syntenic during evolution⁵. In addition, in species closely related in evolution, such as mouse and rat⁷ or man and non-human primates⁸, several homologous genes

have been assigned to chromosomes that are apparently homologous by chromosome banding. Five genes in the mouse (*Eno-1*, *Pgd*, *Pgm-2*, *Ak-2*, *Gpd-1*) are syntenic⁹⁻¹¹ and their human homologues have been assigned to human chromosome 1; all but the human homologue of *Gpd-1* are regionally assigned to arm 1p (refs 4 and 12). This apparent conservation of a rather large autosomal syntenic group prompted us to investigate the extent of conservation of other autosomal regions. The results have provided chromosomal assignments for seven gene loci in the mouse and evidence for synteny of four pairs of gene loci on four different human and mouse autosomes.

Chinese hamster × mouse somatic cell hybrids³¹ segregating mouse chromosomes (independent primary clones) were scored for the expression of murine enzymes including seven for which the gene loci had not yet been assigned in the mouse: PEP-S, PEP-D, LDH-A, ID-2, PK-3, HK-1 and PP. The mouse electrophoretic phenotypes for several of these enzymes are presented in ref. 12.

A potential problem in comparative mapping is the establishment of the homology of genes in different species. The mouse and human forms of PEP-S, LDH-A, PEP-D, PK-3, ID-2 and PP are thought to be homologous because of heteropolymers formed in rodent × human hybrid cells^{4,13}. In man, three hexokinases (HK-1, HK-2, HK-3) have been described¹⁴. Human HK-1 is a monomer, expressed in all tissues and cultured cells and is inhibited by high glucose concentrations. The mouse hexokinase we have scored as HK-1 exhibits all these properties.

The segregation patterns of 29 mouse enzymes scored in 27–40 hybrid clones revealed concordance of enzyme expression or lack of expression between: PEP-S and PGM-1 (*Pgm-1* is assigned to chromosome 5 (refs 11 and 15)); PEP-D, ID-2, and LDH-A with GPI-1 (*Gpi-1* is assigned to chromosome 7 (refs 11 and 15)); PK-3 and MPI-1, MOD-1 (*Mpi-1*, *Mod-1* are assigned to mouse chromosome 9 (refs 11, 15 and 16)); and HK-1 and PP with TRIP-1 (*Trip-1* has been assigned to chromosome 10) (Table 1).

Table 1 Segregation of mouse enzymes in hybrid clones

			PEP-S	LDH-A	PEP-D	ID-2	PK-3	PP	HK-1	
		Mouse			(Number of clones tested)					
Mouse enzyme	McKusick no.	chromosome assignment	31	39	35	27	39	31	28	
					% Discordancy					Refs
DIP-1 (PEP-C)	17000	1	34	18	17	7	36	26	32	15, 17
ID-1 (IDH-1)	14770	1	48	23	26	23	33	18	22	11,15
AK-1	10300	2	32	24	20	19	32	20	26	15
PGD (6PGD)	17220	4	34	33	31	26	31	19	21	9, 15
PGM-2 (PGM-1)	17190	4	37	31	29	26	28	19	21	15, 17
ENO-1	17245	4	34	33	31	26	31	19	21	10
AK-2	10302	4	41	36	33	33	28	24	27	10
PGM-1 (PGM-2)	17200	5	3	30	30	30	21	38	46	11, 15
PEP-S	17020	(5)	—	31	29	27	26	41	44	Reported here
TPI-1	19045	6	29	39	33	38	26	25	26	18
GPI-1	17240	7	31	0	0	4	33	23	29	11, 15
PEP-D	17010	(7)	29	0	—	0	31	20	26	Reported here
LDH-A	15000	(7)	31	—	0	4	33	23	29	Reported here
ID-2 (IDH-M)	14765	(7)	27	4	0	—	33	19	26	Reported here
APRT	10260	8	21	14	15	15	31	30	33	15, 19
GR-1 (GSR)	13830	8	12	19	16	21	19	28	33	15, 20
MOD-1 (ME-1)	15425	9	23	31	29	30	3	29	29	11, 15
MPI-1	15455	9	26	33	31	33	0	29	29	15, 16
PK-3	17905	(9)	26	33	31	33	—	29	29	Reported here
TRIP-1 (PEP-B)	16990	10	40	28	26	22	31	3	0	15
HK-1	14260	(10)	44	29	26	26	29	0	—	Reported here
PP	17903	(10)	41	23	20	19	33	—	0	Reported here
ACP-1	17150	12	37	15	9	19	33	23	25	15
ES-10 (ES-D)	13328	14	29	41	37	41	38	45	39	7, 15
NP-1 (NP)	16405	14	32	37	36	42	40	47	39	7, 15
GLO-1	13875	17	22	21	22	21	26	35	44	18, 21
DIP-2 (PEP-A)	16980	18	38	42	38	37	36	29	38	15
AGAL (AGAL-A)	30150	X	36	19	13	21	31	35	39	15, 19
HPRT	30800	X	43	21	20	30	44	32	36	15, 19, 22
χ^2			25.6	28.0	23.6	16.7	43.5	24.2	28.0	

The primary independent hybrid clones were derived from fusion of Chinese hamster (*Cricetulus griseus*) E36 cells and *Mus musculus* spleen cells from BALB/c, AKR, NZB, and NIH Swiss mice³¹. Mouse and Chinese hamster enzymes were distinguished by vertical starch gel electrophoresis of cell extracts as before^{7,13,32,33}. Mouse enzyme nomenclature is used where available and human nomenclature is given in parentheses. Otherwise human enzyme nomenclature⁴ is used. We have attempted to conform to the recommendations of the mouse nomenclature committee³⁴ in establishing gene symbols. The McKusick catalogue numbers³⁵ are provided which we suggest be used as a common reference for homologous genes in different species. The percentage of clones showing discordant phenotypes for the enzymes (+/–, –/+) are given. A-GAL, α galactosidase; ACP-1, red cell acid phosphatase –1; AK-1, AK-2, adenylate kinase-1 and -2; APRT, adenine phosphoribosyl transferase; ENO-1, enolase-1; ES-10, (human ES-D) esterase-10; GLO-1, glyoxylase-1; GPI-1, glucose phosphate isomerase-1; glutathione reductase-1, GR-1 (human GSR); hypoxanthine phosphoribosyl transferase, HPRT; HK-1, hexokinase-1; ID-1, (human IDH-1) isocitrate dehydrogenase-1 (soluble form); ID-2 (human IDH-M) isocitrate dehydrogenase-2 (mitochondrial form); LDH-A, lactate dehydrogenase A; MOD-1 (human ME-1) malic enzyme soluble form; MPI-1, mannose phosphate isomerase-1; NP-1, purinet nucleoside phosphorylase-1; DIP-1 (human PEP-C) dipeptidase-1; DIP-2, (human PEP-A) dipeptidase-2; TPI-1, triosephosphate isomerase-1; TRIP-1, (human PEP-B) tripeptidase-1; PEP-D, peptidase D; PEP-S, peptidase S; PGD (human 6PGD), 6-phosphogluconate dehydrogenase; PGM-1, (human PGM-2) phosphoglucomutase-1; PGM-2 (human PGM-1), phosphoglucomutase-2; PK-3, pyruvate kinase-3 (also called PK_{M2} in human nomenclature); PP, inorganic pyrophosphatase. The chromosome assignments in parentheses are derived from the present work. In addition, four clones were back selected in 6-thioguanine and PEP-S, LDH-A, PEP-D, ID-2, PK-3, PP, HK-1 segregated independently of the X-linked markers, HPRT and AGAL. The χ^2 values are derived from 2 × 2 comparisons of PEP-S with PGM-1; LDH-A, PEP-D, ID-2 with GPI-1; PK-3 with MPI-1 and MOD-1; HK-1 and PP with TRIP-1. They are all highly significant.

Chromosomes were analysed using Hoechst 33258 staining²³ and trypsin-Giemsa banding methods^{15,24} in more than 30 clones. Those with multiple chromosome rearrangements or without recognisable mouse chromosomes were excluded from further analysis. The 24 clones selected for analysis had largely intact mouse chromosomes. The scoring of mouse chromosomes in the hybrid cells had been validated by the correct hybrid cell chromosome assignment of genes for 11 enzymes assigned by sexual genetics¹⁵. The chromosome data summarised in Table 2 assign *Pep-S* to chromosome 5, *Pep-D*, *Ldh-A* and *Id-2* to chromosome 7, *Pk-3* to chromosome 9 and *Hk-1* and *Pp* to chromosome 10.

Comparison of the mouse and human gene maps using data presented here and previously indicates that several mouse and human chromosomes carry homologous pairs of genes: mouse 4 (*Pgm-2*, *Ak-2*, *Eno-1*, *Pgd*) and human 1p; mouse 5 (*Pgm-1*, *Pep-S*, *Alb-1*²⁵) and human 4; mouse 5 (*Gus*, *Mor-1*)²⁶ and human 7; mouse 7 (*Gpi-1*, *Pep-D*) and human 19; mouse 7 (*Ldh-A*, *Hbb*, mouse beta chain of haemoglobin²⁷) and human 11²⁸; mouse 9 (*Mpi-1*, *Pk-3*) and human 15q; mouse 10 (*Hk-1*, *Pp*) and human 10; mouse 11 (*Glk*, *Tk-1*)²⁹ and human 17q; mouse 17 (*H-2*, *Glo-1*)^{18,21} and human 6p; and mouse X

(*Pgk*, *Agal*, *Hprt*, *G6pd*^{4,8}) and human Xq. (References for the human gene assignments are summarised in refs 4, 12 and 13.) The recombinational distance for many of these gene pairs is unknown in either species and indicates the need for complementary studies by somatic cell genetics and sexual genetics. The known distances suggest that considerable segments are conserved. In the mouse, *Pgm-2* and *Pgd* are 23 map units apart⁹, and in the human male the recombination fraction between PGM and PGD is 0.50¹². The distance between *Gus* and *Mor-1* is 11 map units in the mouse²⁶; in both mouse²¹ and the human male³⁰ *Glo-1* and the major histocompatibility complex (*H-2* or *HLA*) are 5 map units apart.

The evolutionary significance of preservation or disruption of synteny groups is not known. Numerous chromosome rearrangements occurred during the divergent evolution of primates and rodents, resulting in quite different chromosome banding patterns. Therefore each mouse and each human autosome is expected to carry some genes or syntenic groups that are on several different chromosomes in the other species, for example, one part of mouse 5 may be homologous to human 4 and another part to human 7. It has been suggested that during vertebrate evolution, chromosomal rearrangements which tended to preserve ancestral synteny groups have been favoured pathways for the development of new species³⁷. Our data support such hypotheses in part and suggest that comparative gene mapping will add to the understanding of the evolution of the mammalian genome. Similarly, it will be interesting to compare mouse and human developmental defects with mutations, chromosome deletions or additions in regions shown to be homologous by gene mapping.

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Table 2 Segregation of mouse enzymes and chromosomes in hybrid clones

Chromosome	PEP-S	LDH-A	PEP-D	ID-2	PK-3	PP	HK-1
	19	24	No. of clones tested			19	18
			22	17	23		
			% Discordant clones				
1	50	17	18	12	42	24	26
2	32	21	18	24	29	29	26
3	38	35	29	31	35	25	22
4	43	43	43	44	35	30	28
5	0	38	40	43	33	47	53
6	27	42	36	41	25	29	26
7	41	0	0	0	42	19	26
8	24	26	29	31	30	40	39
9	24	43	48	44	0	40	33
10	52	23	20	20	32	5	0
11	55	88	91	82	54	71	63
12	50	17	9	18	42	24	26
13	50	25	27	35	42	38	37
14	37	57	53	67	38	37	29
15	45	13	9	18	46	29	37
16	32	25	18	29	42	24	26
17	27	21	23	24	29	43	47
18	36	38	36	35	38	33	37
19	32	29	27	24	29	43	42
X	45	13	9	18	46	29	37
Y	57	78	76	71	57	55	50
χ^2	18.1	10.7	6.48	8.83	23.6	10.9	15.3

Standardised mouse chromosome nomenclature is used^{34,36}. Between 11 and 25 metaphases per clone were karyotyped and scored for mouse chromosome content on cultures prepared simultaneously for enzyme analysis when the enzyme results were not known. Clones were scored as positive (+) for a given chromosome if its frequency was greater than 0.15 while those below 0.05 were scored as negative (-). When a clone had a particular chromosome with a frequency between 0.15 and 0.05 that chromosome in that clone was not used for analysis. Occasionally, mouse chromosomes were involved in centric fusion (Robertsonian) translocations but were scored as if present independently. In the 24 clones used for analysis there was minimal rearrangement of mouse chromosomes: seven insertions, three deletions (both of the X chromosome), one marker, one Rb(9; 18) one t(9; 6). Discordant clones are of the classes chromosome/enzyme: +/— or —/+. The χ^2 values are derived from 2×2 comparisons of PEP-S with chromosome 4; LDH-A, PEP-D, ID-2 with chromosome 7; PK-3 with chromosome 9; PP and HK-1 with chromosome 10. They are all highly significant. One clone had mouse PP activity but lacked mouse TRIP-1 activity and an identifiable chromosome 10.

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Deletion mapping of the *t* complex of chromosome 17 of the mouse

THE dominant *T* mutation in the *t* complex on chromosome 17 of the mouse causes a shortening of the tail in heterozygotes and embryonic lethality in homozygotes¹. Recessive *t* alleles are defined by their interaction with *T* to cause tail-lessness in double heterozygotes (*T/t*) and are often associated with three other properties: (1) homozygous lethality at several different embryonic stages, (2) effects on male reproduction including sterility and transmission ratio distortion, and (3) crossover suppression^{2,3}. Although the multiplicity of these effects suggests that several genes are involved, crossover suppression has made classical genetic analysis of the region difficult. As *T/t* animals are viable, the lethal factors associated with recessive *t* homozygosity cannot be allelic to those associated with *T* homozygosity. Lyon *et al.* studied exceptional crossovers and found evidence that the homozygous lethality of *t* alleles and their property of interacting with *T* to cause tail-lessness were specified at different genetic loci^{4,5}. We have studied a presumed deletion, *T^{Orl}*, in crosses with recessive *t* alleles as well as with alleles at closely linked loci to map further the multiple effects associated with the *t* complex. We report here that factors associated with *t* lethality are distal to the *T^{Orl}* deletion and separable from those responsible for at least one kind of *t* allele-associated male sterility apparently included in the deletion.

Moutier suggested in a preliminary report⁶ that *T^{Orl}* was a deletion and extended as far as quaking, as the latter, recessive mutation was expressed in a heterozygote (*trans*) with *T^{Orl}*, due to hemizyosity. We have confirmed this (Fig. 1): all the short-tailed offspring (*T^{Orl}/qk*) from *qk/qk* × *T^{Orl}/+* crosses expressed the quaking phenotype but none of the normal-tailed mice showed the quaking phenotype. Furthermore, *T^{Orl}/qk* males were sterile and their vasa deferentia contained no sperm, a characteristic of male *qk* homozygotes.

The fused-kink locus and the tufted locus are, respectively, two and three centimorgans distal to *qk* (Fig. 1). Bennett has reported that the deletion of *T^{Orl}* does not extend to the recessive marker *tf* (ref. 7). We have crossed heterozygotes for *T^{Orl}* with heterozygotes for *Fu^{ki}*, which is also lethal when homozygous⁸. Of 76 offspring, 25 had tails characteristic for *T^{Orl}*, 19 had tails characteristic for *Fu^{ki}* (there is some overlap between the phenotypes, so these classifications are not absolute), 12 had a tail-less phenotype, and 20 had normal tails. The 3:1 segregation of abnormal tails to normal tails suggests that *Fu^{ki}/T^{Orl}* compound heterozygotes are viable and have abnormal tails. From the apparent viability of double heterozygotes we conclude that the deletion represented by *T^{Orl}* does not extend as far as *Fu^{ki}* and terminates somewhere between quaking and fused-kink.

In the other direction on chromosome 17, the results of crosses of *T* with *T^{Orl}* strongly suggest that *T^{Orl}* acts as an allele of *T* with regard to developmental lethality. Examination of embryos from *+/T^{Orl}* × *+/T^{Orl}* matings revealed that the

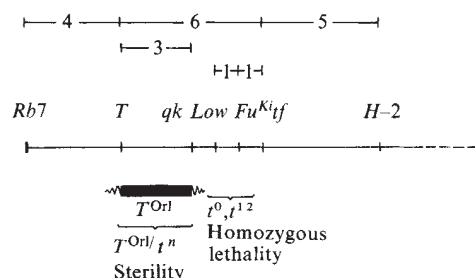
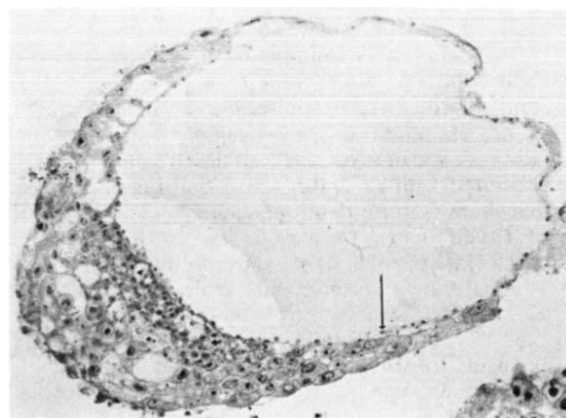


Fig. 1 The proximal portion of chromosome 17 of the mouse and the location of *T^{Orl}*-mapped *t* factors. Above, linkage distances (from refs 14-16); centre, the chromosomal map; below, the location of the *T^{Orl}* deletion and new assignments (bracketed) of *t* allele effects from this paper. *qk*, quaking, *Fu^{ki}*, fused-kink, *tf*, tufted.

T^{Orl}/T^{Orl} progeny are similar in many ways to lethal triploid⁹ and *T^{Orl}* homozygous⁷ embryos, although *T^{Orl}/T^{Orl}* embryos are more retarded in growth and seem to have a somewhat earlier lethal period than either of these (Fig. 2). These *T^{Orl}/T^{Orl}* homozygotes die before 10 d of development and bear no resemblance to *T/T*. However, in studies of embryos from *+/T^{Orl}* × *+/T* matings we found that the double heterozygote *T/T^{Orl}* resembled *T/T* lethal embryos. These data indicate that the *T^{Orl}* deletion extends to *T*.

The interaction of *T^{Orl}* with recessive lethal *t* mutations belonging to two different complementation groups was also studied. *t¹²* and *t⁰*, when homozygous, result in developmental arrest at the morulae and egg cylinder stages, respectively². Crosses of *+/t¹²* as well as *+/t⁰* with *T^{Orl}/+* resulted in viable tail-less offspring. These must be *T^{Orl}/t¹²* and *T^{Orl}/t⁰* rather than highly expressed (tail-less) *T^{Orl}/+* because (1) crosses of putative *T^{Orl}/t¹²* × *T^{Orl}/t¹²* mice only resulted in tail-less offspring, in very small numbers (that is, the intercross creates a balanced lethal line where only double heterozygotes survive), and (2) the tail-less males are quasi-sterile but *T^{Orl}/+* males are not (see below). Furthermore, we have cultured 41 3½-day-old embryos from *+/t¹²* × *T^{Orl}* matings for one day and found only 2% of the embryos arrested developmentally as morulae. This is significantly less than expected if the *T^{Orl}/t¹²* progeny died at this stage ($\chi^2 = 8.4$, $P < 0.005$). Thus, development of *t¹²/T^{Orl}* embryos cannot cease at the morula stage. As *tⁿ* lethality is recessive, hemizyosity for *tⁿ* would be expected to be lethal or nearly so. Thus, factors determining *t⁰* and *t¹²*

Fig. 2 Histological appearance of *T^{Orl}/T^{Orl}* embryos at 8.5 d of development showing features similar to those seen in triploid embryos. These lack any embryonic development and consist only of an inner parietal endoderm layer (arrow) surrounded by trophoblastic giant cells. Longitudinal, 7-μm section stained with haematoxylin and eosin. Magnification ×1,500.



embryonic lethality are likely to be located distal to the termination of the T^{Orl} deletion as depicted in Fig. 1. These results agree with those of Lyon and Mason¹⁰ which indicate that factors associated with recessive t alleles map near tf , which is also outside the T^{Orl} deletion.

Lethal t alleles affecting different stages of embryonic lethality complement each other at least partially, although complementing, doubly heterozygous males are sterile. To determine if a portion of the genome responsible for male sterility is included in the chromosomal region deleted in T^{Orl} , we tested the fertility of several T^{Orl}/t^{12} and T^{Orl}/t^0 males and found it to be markedly reduced in males of both genotypes (Table 1). Although a few offspring were born to such males, their fertility was less than 5% of normal and they are thus quasi-sterile. Thus, T^{Orl} complements t^{12} and t^0 for lethality but not for full fertility. Lyon and Mason¹⁰ defined one class of sterility caused by genes in the t complex as the interaction of t alleles derived from t^6 with t^{w5} to cause sterility. This sterility effect was inseparable by recombination from t allele lethality. In contrast, our data show a dissociation of at least one kind of sterility from lethality in the t complex. In addition, it is possible that factors affecting male t allele transmission ratio distortion are also located in the chromosomal region deleted by T^{Orl} . T^{Orl} shows a slight segregation distortion in males, 220/180, $\chi^2 = 4.0$, $P < 0.05$.

Table 1 Fertility of T^{Orl} in combination with various alleles compared to controls

Male genotype	No. tested	No. female* weeks per male	Newborns per female per week†
$T^{Orl}/+$	3	15	5.61
T^{Orl}/t^{12}	4	15	0.08
T/t^{12}	2	18.5	5.65
T^{Orl}/t^0	3	14	0.28
T/t^0	5	27	7.85

* The indicated males were caged with mature, virgin, random-bred CD₁ females of unproven fertility which were replaced at the end of each 7-d period.

† The separated females were examined twice a week for newborns which were counted and removed.

In summary, we suggest that T^{Orl} is a deletion extending from T to some point distal to qk , but not as far as Fu^{ki} . Furthermore, the developmental lethal effects of t^{12} and t^0 apparently map distal to the end of the deletion. Lyon and Bechtol¹¹ have presented evidence that a t^6 -derived t allele, t^{20} , is a deletion in the region of tf . As this deletion allows the expression of other t allele recessive lethal factors, a location near tufted is likely.

Thus, quaking is mapped well within the t complex. Quaking is a neurological mutant which is male sterile. Morphological studies have shown marked similarities in the distortion of sperm head shape found at the late spermatid stage in qk/qk (ref. 12) and in mice homozygous for a semi-lethal, male sterile t allele.¹³ Furthermore, at least one exceptional crossover from t^6 was neurologically abnormal⁵. Thus, the phenotypic expression of qk may be another example of the spectrum of effects that are induced by mutations in the t complex.

Lyon and Mason¹⁰ have presented arguments for the separation of an 'abnormal ratio' region from a 'sterility' (defined only as sterility of t^n/t^{w5} males) region which was closely associated with an embryonic lethal region. Our data suggest that there is more than one cause of sterility and that sterility of T^{Orl}/t^n males is separable from t^n/t^n lethality and could be associated with qk/qk sterility. Furthermore, this sterility is apparently associated with male transmission ratio distortion. There is also more than one region in this part of chromosome 17 controlling embryonic development. Thus, the t complex seems to provide an example of a 'supergene'—a large chromosomal segment with multiple genes involved in similar, or closely related functions. The nearby $H-2$ region is,

of course, another example of this phenomenon and the proximity of the two supergenes has often raised the question of whether the multiple functions of these two regions could be related.

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Simian virus 40 T antigen binds to host cell chromosomes

T ANTIGEN of simian virus 40 (SV40) is the product of the A gene of the virus¹ and has been shown to have a role in the establishment and maintenance of transformation^{2–5}. T antigen has also been shown to be required for the initiation of each round of viral DNA synthesis⁶ and for induction of host DNA synthesis^{7–11}. Furthermore, it has been suggested that T antigen may initiate DNA synthesis in transformed cells and thereby help maintain those cells in the transformed state^{6,12–14}. The SV40 genome has three preferential binding sites for T antigen, one of which is at or near the site at which viral DNA synthesis is initiated¹⁵. The binding of T antigen to eukaryote DNA is less well defined but shows a preference for double-stranded over single-stranded DNA¹⁶. Similar findings have been reported for tumour-specific transplantation antigen¹⁷. SV40 nucleohistone has also been reported to bind T antigen in amounts threefold greater than does native SV40 DNA¹⁸. To investigate the role of T antigen in the regulation of the host cell, we have asked whether host chromosomes bind detectable amounts of T antigen; and, if so, whether the binding is restricted to specific chromosomes or to specific chromosomal loci. Although earlier studies have suggested that T antigen is associated with nuclear chromatin of infected cells¹⁹, this study is the first to report the binding of SV40 T antigen to mammalian chromosomes.

We have used a fibroblastic cell strain derived from a male Indian deer, *Muntiacus muntjak*²⁰. This cell strain can be infected by SV40 virus as shown by the nuclear accumulation of T antigen, and the induction of host DNA synthesis; neither capsid protein nor infectious progeny are produced (E.L.G., unpublished). Our choice of cell strain was dictated by the fact that the male muntjac has only seven chromosomes, which can be unequivocally identified. Karyotype analysis is easily performed in such cells and the T antigen binding sites readily identified.

Cultures were grown in Eagle's medium supplemented with foetal bovine serum 10% (Gibco), potassium penicillin G (500,000 units l⁻¹), streptomycin sulfate (100 mg l⁻¹), and mycostatin (25,000 units l⁻¹). Cultures were maintained at a density of 3×10^3 to 8×10^4 cells cm⁻² by weekly passage in plastic roller bottles (Corning) or 75 cm² flasks (Falcon). All experiments were performed with cells in their 20th to 35th generation. Cells seemed to be free of mycoplasma by fluorescence staining with Hoechst 33258²¹ and by scanning electron

microscopy²². The virus used for these experiments was strain VA 4554 grown in a subclone of the CV-1 cell line with infection at low input multiplicity as described previously²³. Stock virus had a titre of 10^9 plaque-forming units (PFU) per ml and an equivalent number of T antigen-forming units per ml. Muntjac cells were infected at multiplicities between 200 and 500 PFU per cell. After 2 h adsorption, incubation was continued at 37°C for 48 to 72 h before the selection and/or fixation of mitotic cells. Mitotic cells were selectively detached from logarithmically growing roller cultures²⁴, washed once in phosphate-buffered saline (PBS), swollen in 0.075 M KCl for 12 min at 37°C, fixed in suspension with methanol-acetic acid (3:1) for 15 min, dropped on to wet, frozen, glass slides, and air dried²⁵. Alternatively, cells were grown on glass slides pre-cleaned with acetic acid and ethanol; these cells were arrested with $0.25 \mu\text{g ml}^{-1}$ colcemid (Gibco) for 4 h, swollen as above and fixed *in situ* with methanol-acetic acid (3:1) for 15 min²⁶. Although many more metaphases were obtained by selection of mitotic cells, fixation *in situ* is more gentle; no differences in chromosome morphology were detected with these two methods of preparation. The solubility of T antigen did not seem to be affected by colcemid arrest, hypotonic swelling, or fixation as judged by comparison of fluorescence intensity and end-point titrations by the indirect immunofluorescence technique²⁷ with both muntjac and CV-1 cells. Chromosome preparations were rehydrated with PBS and then incubated at

37°C for 2 h with a 1:20 or 1:50 dilution of hamster anti-T serum (Flow Labs or NCI) which had been previously adsorbed on monolayers of both muntjac and CV-1 cells. After washing with PBS, preparations were incubated at 37°C for 2 h with a 1:10 dilution of fluorescein isothiocyanate-conjugated (FITC) goat anti-hamster IgG immunoglobulin (Antibodies Inc.).

Chromosome preparations made from infected cultures were brightly stained (Fig. 1), however, not all metaphase spreads from infected cultures were fluorescent. This correlates well with the fact that 25% of the nuclei in cultures inoculated with SV40 were not fluorescent. All chromosomes in an infected diploid or an occasional tetraploid cell appeared to have bound T antigen. Furthermore, the T antigen was not evenly distributed along the chromosomes, as evidenced by the intense fluorescent regions or bands, which appear in a similar pattern for all chromosome complements. Chromosomes from uninfected cultures were not fluorescent; in fact it was not possible to obtain a photograph of these chromosomes with the low background fluorescence. The substitution of normal hamster serum for hamster anti-T serum in the staining procedure did not result in the appearance of any T antigen positive nuclei or chromosomes.

The chromosomes of SV40-infected cells fluoresce when stained with antisera that react with T antigen by complement fixation. In the absence of purified T antigen with which to inhibit the binding of anti-T serum, we have used normal hamster serum to limit the possibility that the binding of the FITC-conjugated goat anti-hamster IgG immunoglobulin is due to something in the hamster serum other than antibody to T antigen. This possibility is further diminished since the same results were obtained with two different sources of anti-T serum. The fact that uninfected cells do not contain fluorescent chromosomes and presumably uninfected cells within the SV40-treated population also do not fluoresce, suggests that the binding of T antigen is specific for the chromosomes of infected cells and is not an artefact of the fixation and staining procedure. While it is possible that the pattern of T antigen banding that we see is artefactual and generated by a relocation of the T antigen during swelling and fixation, a similar banding pattern has been observed for at least 500 chromosome complements derived from four different infected cell populations and two different hamster anti-T sera. It is also possible that an alteration of the structure of the chromosomes has been caused by SV40 infection and that we are observing a banding pattern which is a consequence of the rearrangement of other, non-virally coded, chromosomal proteins, which nonetheless affect the binding of T antigen. This seems unlikely however, since without previous trypsin treatment chromosomes from SV40 infected cells show no banding pattern when stained with Giemsa. Comparison of the T antigen patterns with those obtained by Q and G banding techniques (to be published in detail elsewhere) indicates that these established banding patterns are not altered by SV40 infection and that the T antigen pattern is very similar. Therefore, while it is possible that T antigen binds to initiation sites for host cell DNA replication, it is unlikely that it binds to sites at which SV40 DNA has integrated. In fact, it has been reported that T antigen binding is not restricted to the origin of SV40 replication and that the SV40 nucleohistone complex binds three times more T antigen than does the native SV40 DNA^{15,18}. Beyond the initiation of viral DNA synthesis, the function of the T antigen is unclear. It remains to be seen whether T antigen is serving a structural or functional role in this instance or whether its pattern of binding is simply a passive result of the chromosome substructure. From this approach we hope to learn more about the function of T antigen and the viral regulation of the host cell.

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Fig. 1 Muntjac chromosomes swollen in 0.075 M KCl for 12 min, fixed in 3:1 methanol:acetic acid, air dried on slide, and stained for T antigen by indirect immunofluorescence 72 h after infection with SV40 were photographed on an inverted microscope equipped for epifluorescent excitation at 495 nm and suppression at 525 nm (Leitz). The photograph was taken on Tri-X film with automatic metering at 32 DIN at a magnification of 252 and is shown here at a magnification of $\times 1285$. T antigen is associated with all chromosomes, including the small Y, and appears as bright fluorescent bands.



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Influence of the major histocompatibility complex on lymphocyte interactions in antibody formation

PRODUCTS of the major histocompatibility gene complex (MHC) play an important part in regulating immune responses, particularly responses involving the participation of thymus-derived (T) lymphocytes. It seems that wherever the situation has been carefully examined, T cells exhibit a dual specificity; that is, specificity for, on the one hand, structures expressed by the MHC and, on the other, for foreign antigenic determinants. Thus, in the mouse, cytotoxic T cells show specificity for H-2K or H-2D (refs 1, 2) determinants, whereas those involved in delayed type hypersensitivity³ and those which help in antibody formation⁴ show specificity for the I region. The nature of MHC restrictions exhibited by cytotoxic cells has been much clarified by the studies of Bevan⁵ which suggest, and of those of Zinkernagel *et al.*⁶, which show, that precursors of cytotoxic T cells are probably selected for their capacity to recognise particular H-2 markers during their differentiation in the thymus, and that this 'learning' process is not influenced by the H-2 haplotype of the T cells themselves. The data presented here suggest that a comparable situation exists for T lymphocytes (helper T cells) participating in antibody production.

The realisation that MHC gene products are crucial to T-B lymphocyte cooperation came with the findings of Kindred and Shreffler⁷ and of Katz *et al.*⁸ that histoincompatible T and B cells cooperate poorly, if at all. The restricting loci in this system seemed⁹ to be located in the I-A subregion of H-2. This

phenomenon was initially thought to involve obligatory interactions between like determinants on the cooperating cells. However, this hypothesis lost plausibility when it was demonstrated^{10,11} that irradiated bone marrow reconstituted mouse chimaeras of the type $P+Q \rightarrow (P \times Q)F_1$ could be made to generate populations of T helper cells of P or Q origin, both capable of cooperation with either P or Q B-cells (P and Q are used here purely as symbols). In such double chimaeras the lymphoid system consists of roughly equal numbers of parental lymphocytes, with very few lymphocytes of host origin. The issue now arose as to whether this apparent lack of MHC restriction reflected the elimination of allogeneic inhibitory elements (see below) from the P and Q lymphocyte populations of the chimaeras, or whether some form of adaptive differentiation had occurred such that the T cells of each parental type had learnt to regard the other haplotype as 'self' and that helper T-cell precursors with appropriate 'learnt-self' specificities had been selected. Indeed studies of Sprent and von Boehmer¹² suggested that the latter explanation was most likely.

Table 1 Helper activity of T cells depleted of alloreactivity

Source of T cells primed to KLH		Total PFC with the following B cells	
		CBA (H-2 ^k)	BALB/C (H-2 ^d)
Expt 1 Single chimaeras*			
C3H/He $\rightarrow$ (BALB/c $\times$ C3H/He) _F ₁		1,163	20
[(H-2 ^k) $\rightarrow$ (H-2 ^d $\times$ H-2 ^k) _F ₁]			
BALB/c $\rightarrow$ (BALB/c $\times$ C3H/He) _F ₁		18	1,733
None		50	78
Expt 2a Neonatally tolerant donors†			
CBA (H-2 ^k) tolerant of BALB/c. Donors		3,652	0
1 + 2 (pool)			
(BALB/c $\times$ C3H/He) _F ₁ Control		3,100	1,974
None		0	0
Expt 2b Donors 3		72	0
4		428	55
BALB/c control		NT	6,902
CBA control		662	NT
None		92	36
Expt 2c Donors 5		501	0
6		536	6
BALB/c control		NT	3,440
CBA control		1,602	63
None		39	0

Mice of various haplotypes (as indicated in the left column) were primed with KLH in Freund's complete adjuvant (FCA). T cells were purified by the passage of cells through nylon wool. Red cells were removed by treatment with ammonium chloride and the remaining T cells were treated at 37°C for 20 min with 25 μ g ml⁻¹ of mitomycin C to eliminate any potential allogeneic inhibitor cells.¹⁵ The cells were then assessed for helper function by incubation for 5 days with either BALB/c or CBA TNP-OVA primed spleen cells ('B cells') which had been treated with anti-Thy-1 serum and complement, and antigen (0.1-3 μ g ml⁻¹ of TNP-KLH). Direct and indirect plaque-forming cells (PFC) to TNP-coupled donkey erythrocytes were counted in 30 micro-culture wells (10- μ l aliquots) on day 5. The values shown represent the total number of plaques in the 30 wells.

* Single chimaeras were constructed by injecting 5 $\times$ 10⁶ anti-Thy-1 and complement treated bone marrow cells from either C3H/He or BALB/c donors into adult, irradiated (875 rad) (C3H/He $\times$ BALB/c)_F₁ recipients. Chimaerism was confirmed in randomly selected mice with cytotoxic antisera. Pooled spleens from pairs of mice were used for each group. Similar results were obtained in many comparable experiments.

† CBA mice were made tolerant to BALB/c histocompatibility antigens by neonatal i.v. administration of 25 $\times$ 10⁶ (C3H/He $\times$ BALB/c)_F₁ spleen cells²¹. All mice were grafted with BALB/c tail skin 7 weeks after birth²², and mice with healthy grafts after a further 2 months were used in subsequent experiments. Complete absence of allograft rejection was confirmed histologically.

Table 2 Relevance of priming environment in the generation of T helper cells

Source of T cells	MHC haplotype of priming environment	Total PFC with following B cells	
		CBA ($H-2^k$)	BALB/c ($H-2^d$)
Expt 1*			
BALB/c LN cells from (BALB/c × C3H/He) double chimaeras	$H-2^d$	536	2,995
	$(H-2^d \times H-2^k)F_1$	5,148	2,846
None		144	443
Expt 2*			
BALB/c cells from BALB/c → (BALB/c × C3H/He) F_1 single chimaeras	$H-2^d$	839	2,392
	$(H-2^d \times H-2^k)F_1$	3,734	250
None		0	0
Expt 3†			
C3H/He cells from C3H/He → (BALB/c × C3H/He) F_1 single chimaeras	$H-2^k$	2,875	313
	$(H-2^d \times H-2^k)F_1$	259	1,283
None		12	5
Expt 4‡			
CBA tolerant to BALB/c	$H-2^k$	489	0
	$(H-2^d \times H-2^k)F_1$	1,220	13
None		34	0
BALB/c		NT	655

All cell recipients were primed with 500 µg KLH in FCA, and 1 week later their spleen T cells were prepared, pooled, purified and tested as described in Table 1. The figures represent the total number of anti-TNP PFC in 30 wells. All experiments have been repeated with comparable results.

* Aliquots of 25×10^6 lymph node cells from 8 BALB/c × C3H/He → (BALB/c × C3H/He) F_1 double chimaeras (Expt 1) or BALB/c → (BALB/c × C3H/He) F_1 single chimaeras (Expt 2) were transferred into either 6 BALB/c ($H-2^d$) or 6 (C3H/He × BALB/c) F_1 [that is ($H-2^k \times H-2^d$) F_1] recipients irradiated with 600 rad. All cell populations were treated before transfer with anti- $H-2^k$ serum and complement.

† Spleen and lymph node cells from 8 C3H/He → (C3H/He × BALB/c) F_1 chimaeras were transferred into 4 C3H/He or 4 (C3H/He × BALB/c) F_1 recipients irradiated with 750 rad (Expt 3).

‡ Aliquots of 100×10^6 spleen and lymph node cells from 4 CBA donors rendered neonatally tolerant to BALB/c were transferred into 4 CBA or 4 (CBA × BALB/c) F_1 recipients irradiated with 750 rad.

One of the problems of culturing histoincompatible T and B lymphocytes is that certain T cells are generated which will actively inhibit antibody formation. However, inhibitory effects of this kind are expressed predominantly when there are *K* or *D* end differences of the $H-2$ region between the interacting cells, and do not therefore explain why *I-A* incompatible cells fail to cooperate^{13,14}. Inhibitory allogeneic reactions which might easily confuse the interpretation of *in vitro* experiments, can be prevented by (1) irradiation¹³ or mitomycin C treatment¹⁵ of the helper T-cells, or by (2) use of helper T cells depleted of alloreactivity. The latter approach can be explored by the induction of classical tolerance in neonatal mice¹⁶ or by the construction of adult chimaeras. In this study a combination of these methods was used, in as much as depletion of alloreactivity by one means or another was followed by treatment of the T cells with mitomycin C as an extra safeguard. The cells were then examined for their ability to provide help in the *in vitro* response of syngeneic and allogeneic B cells to TNP-KLH. In all categories of 'tolerant' mice used, spleen cells were tested for their capacity to mediate allogeneic inhibitory reactions¹³, and were also found to be uniformly tolerant in this respect (data not shown).

Our initial experiments were designed to determine whether antigen-primed T cells from allogeneic donors could cooperate with B cells when alloreactivity had been eliminated. The data show (Table 1) that tolerance between interacting T and B cell populations, be it induced neonatally or in bone marrow chimaeras, cannot guarantee cooperation. Thus, primed T cells from single chimaeras of the type $P \rightarrow (P \times Q)F_1$ or from *P* mice rendered neonatally tolerant of *Q* were found to be extremely inefficient in their capacity to cooperate with cells from the allogeneic strain (*Q*) although fully competent to help $H-2$ compatible (*P*) B-lymphocytes. Single chimaeras constructed from different strain combinations gave essentially similar results¹⁴.

The data obtained with T cells from neonatally injected tolerant mice confirm previous *in vivo* experiments¹⁷ which

also showed persistence of $H-2$ restriction despite mutual tolerance between T and B cells. However, none of the data given in Table 1 are, on first sight, reconcilable with other *in vitro* studies^{16,18} that demonstrate unrestricted cooperation after removal of inhibitory T cells. A possible explanation is, nevertheless, put forward below.

The failure of T cells from *P* mice tolerant of *Q* histocompatibility antigens to cooperate with *Q* B-cells seems surprising when contrasted with the successful cooperation seen when *P* T-cells are derived from antigen-primed double chimaeras $P + Q \rightarrow (P \times Q)F_1$. Thus, the work of Kappler Marrack¹⁹ and of Sprent²⁰ has shown that, for T cells from $(P \times Q)F_1$ hybrids, the MHC haplotype of the initial priming environment determines the MHC preference expressed by antigen-primed cells. We therefore tested the underlying premise that the presence of *Q* MHC determinants in the priming environment was necessary to generate T cells of *P* origin capable of cooperating with *Q* B-cells. When *P* type cells from either $P + Q \rightarrow (P \times Q)F_1$ or from $P \rightarrow (P \times Q)F_1$ mice were adoptively transferred and primed for the first time in an environment containing *Q* (that is, in F_1 hybrids), cooperation with *Q* B-cells was easily demonstrated (Table 2). In contrast, *P* T-cells from mice rendered neonatally tolerant to *Q* could not be primed to cooperate efficiently with *Q* B-cells, regardless of the MHC haplotype of the priming environment. (We have as yet no explanation to account for the fact that cells from single chimaeras cooperate relatively poorly with syngeneic B cells after priming in the F_1 hybrid.) These results lead to the following conclusions. First, the MHC of the priming environment is important in determining the pattern of MHC specificities recognised by T helper cells. Second, this is, in itself, not sufficient, because *P* T-cells from neonatally tolerant mice could not be primed to produce significant levels of cooperation with *Q* B-cells. There is, clearly, a difference between the T-cell populations derived from bone marrow chimaeras and those obtained from neonatally tolerant donors. The experiments described below show that this difference is related to

Table 3 Helper activity of T lymphocytes from $F_1 \rightarrow$ parent chimaeras

Source of T cells		Total PFC with the following B cells	
		CBA ($H-2^k$)	BALB/c ($H-2^d$)
Expt 1			
(BALB/c $\times$ C3H/He) $F_1 \rightarrow$ C3H/He			
Donor 1		2,731	86
2		2,065	108
3		2,825	142
(BALB/c $\times$ C3H/He) F_1 pool		3,181	912
None		108	83
Expt 2			
Donor 4		1,725	0
5		3,131	89
(BALB/c $\times$ C3H/He) F_1 pool		4,184	5,751
None		110	58

$F_1 \rightarrow$ parent chimaeras were constructed by injection of 5×10^6 anti-Thy-1 antibody and complement treated (C3H/He $\times$ BALB/c) F_1 bone marrow cells into irradiated (875r) C3H/He recipients. These mice were primed with KLH and T-cell helper function was then assessed, as before, for BALB/c and CBA B-cells. As before, the figures represent the total number of anti-TNP PFC in 30 wells. Control F_1 T-cells were from a pool of two spleens. Cytotoxicity studies on randomly selected mice demonstrated that no more than 5–10% of host lymphocytes were present in spleens of these $F_1 \rightarrow$ parent chimaeras.

the MHC haplotype of the host in which the bone marrow stem cells differentiate into precursors of helper T cells, and that it is quite unrelated to experimental exposure of the cells to antigen.

It is fundamental to the design of experiments which examine MHC restriction that ($P \times Q$) F_1 T-cells from primed F_1 donors should cooperate with either P or Q B-cells in the response to complex antigens (see Table 1, experiment 2). We therefore sought to determine what, if any, restrictions might be imposed on F_1 cells which had matured in irradiated P and Q recipients. F_1 T-cells would be naturally tolerant of P and Q determinants, and antigen presentation would be performed by cells carrying both P and Q determinants. Table 3 shows data obtained from mice of the type ($H-2^k \times H-2^d$) $F_1 \rightarrow H-2^k$, comparing the help provided by T cells from this kind of chimaera with that of F_1 hybrid T cells. Cells from antigen-primed donors showed clear cooperative preference for B cells identical in MHC haplotype to the host in which the F_1 cells had matured.

Similar restrictions have been seen with ($H-2^b \times H-2^a$) $F_1 \rightarrow H-2^a$ and ($H-2^b \times H-2^a$) $F_1 \rightarrow H-2^b$ chimaeras (unpublished data). However, T cells from one combination, ($H-2^d \times H-2^k$) $F_1 \rightarrow H-2^d$, although demonstrating some overall preference for cooperation with $H-2^d$ compared with $H-2^k$ B-cells, were nevertheless able to cooperate with $H-2^k$ B-cells. In this connection, it is of interest that two previous experiments demonstrating cooperation between T cells and histoincompatible B cells have involved $I-A^d$ T-cells and $I-A^k$ B-cells^{15,18}. It is therefore possible that some unrecognised but potentially interesting one-way cross-reaction between $I-A^d$ and $I-A^k$ gene products makes this combination sometimes amenable to unrestricted cooperation.

Our results comparing cells from adult radiation chimaeras with cells from neonatally injected tolerant donors show that it is the host environment in which T cells differentiate that determines the ultimate $H-2$ restricted specificity of helper T-cells. Tolerance to $H-2$ histocompatibility antigens is insufficient to allow help to be provided to B cells carrying the relevant $H-2$ antigens. It remains to be seen whether that it is the thymus which imprints the cooperation function on helper T cells, as seems to be the case with cytotoxic precursor cells⁶.

There are many possible explanations for MHC restriction of T lymphocytes, but no comprehensive hypothesis has been put forward. The nature of the imprinting mechanism for the

specificities exhibited by helper T lymphocytes and by cytotoxic T cells is likely to provide the key to the solution of this problem and may well clarify the nature and function of MHC-linked I genes, which are also located in the I region.

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Induction of macrophage production and proliferation by a purified colony stimulating factor

COLONY STIMULATING FACTORS (CSFs) stimulate isolated haemopoietic progenitor cells to differentiate and form granulocyte and/or macrophage colonies *in vitro*^{1,2}. There seem to be at least two distinct subclasses of CSF, those stimulating macrophage colony formation and those stimulating granulocytic colony formation. Purified CSF from mouse L cell-conditioned medium is a 70,000 molecular weight glycoprotein, comprising two disulphide-bonded 35,000 MW subunits³. At concentrations as low as 10^{-12} M it stimulates macrophage colony formation by murine bone marrow cells. It has similar physicochemical properties to, and shared antigenic determinants with, the CSF from human urine^{3–6}. This type of CSF has been shown⁶ to share identity with the macrophage growth factor of Virolainen and Defendi⁷ that stimulates proliferation of peritoneal exudate macrophages. The purified CSF was also active⁶ in stimulating macrophage colony formation by peritoneal exudate cells in the agar culture system of Lin and Stewart⁸. We now report that this same kind of CSF is the molecule responsible for the proliferation of mononuclear phagocytes from several haemopoietic and nonhaemopoietic tissues.

Mouse L cell conditioned medium containing 10% fetal calf serum (FCS) was prepared as described previously⁹ using the L60T clonal isolate of mouse L cells¹⁰. Purified mouse L cell CSF (1.5×10^8 units (ref. 11) per mg protein)³, highly purified human urinary CSF (5×10^7 units per mg protein)⁵ as well as

Table 1 Stimulation of macrophage colony formation by purified mouse L cell and human urinary CSFs in cultures of cells from various murine tissues

Source of CSF	Dilution of CSF	Approximate final concentration of CSF (pmol l ⁻¹)	Bone marrow (colonies per 5 × 10 ⁴ cells)	Pulmonary alveolar cells	Blood mononuclear cells*	Non-parenchymal liver cells Colonies per 10 ³ cells	Peritoneal exudate cells	Resident peritoneal cells
None			0	0	0	0	0	0
Mouse L cell-conditioned medium	1		120	222	79	82	82	1
	1/2		138	202	88	88	59	1
	1/4		108	83	59	61	51	0
	1/8		115	27	46	60	32	0
	1/16		91	2	12	44	14	0
	1/32		44	0	0	19	2	0
Purified mouse L cell CSF	1	200	119†	194	87	84	105	1
	1/2	100	107	108	84	90	105	1
	1/4	50	131	61	48	68	92	1
	1/8	25	140	20	45	52	45	0
	1/16	12.5	83	5	28	45	13	0
	1/32	6.25	37	1	0	29	2	0
Purified human urinary CSF	1		130†	68	132	81	82	0
	1/2		149	40	131	56	63	1
	1/4		108	12	75	58	28	0
	1/8		85	2	47	29	23	1
	1/16		51	0	9	9	1	0
	1/32		18	0	0	5	0	0

All CSF samples were diluted as indicated in alpha medium containing 10% FCS and each dilution was present at 10% of the final culture volume. Figures represent the average colony count for duplicate cultures.

* Results expressed as colonies per 10³ cells although there were 3 × 10³ peripheral blood cells per culture.

† Colonies comprising only mononuclear cells.

the rabbit antiserum⁶ to highly purified mouse L cell CSF (approximately 10⁸ units per mg protein) were prepared as described elsewhere^{3,5,6}. The purified mouse L cell CSF used has been shown to be at least 90% pure by each of four independent criteria³.

All cell populations examined were from C3D2F₁ mice (Jackson). Culture systems in which proliferating cells gave rise to colonies were used in order to additionally determine the fraction of cells in each target population that was proliferating in response to CSF. Agar cultures (1.0 ml) of bone marrow cells^{6,12}, and liquid cultures of pulmonary alveolar cells¹³, blood monocytes¹⁴, peritoneal exudate and resident peritoneal cells⁸ were set up using previously reported techniques. Kupffer cell-rich non-parenchymal liver cell suspensions were obtained by a modification of the method of Berg and Boman¹⁵, described in detail elsewhere¹⁶.

The results obtained for cultures of all the cell types examined with the purified L cell and human urinary CSF are summarised in Table 1, together with those for control cultures containing either crude L cell-conditioned medium or no CSF. Purified CSFs from mouse L cell-conditioned medium of human urine possessed the full activity of crude mouse L cell CSF in stimulating these cell populations. The pattern of relative responsiveness of the different cell populations was strikingly similar for each of the three CSF sources. As reported elsewhere, for L cell-conditioned medium⁷ unstimulated peritoneal cells failed to respond to any of the sources of CSF. Proliferation of all of the remaining cell populations had an absolute requirement for the presence of CSF. Bone marrow contained the most sensitive responding cell population, optimal responses being observed at concentrations of purified mouse L cell CSF as low as 25 pmol l⁻¹ (pM). For the other target populations optimal responses were obtained at a significantly higher concentration (~100 pM).

Further evidence that CSF is the only factor in L cell-conditioned medium required for proliferation of these target populations is shown in Table 2. Antiserum to highly purified L cell CSF, when mixed with L cell-conditioned medium in pro-

portions that resulted in 60% inhibition of bone marrow colony formation also inhibited colony formation by the other target populations to a similar degree (63–71%). It has been shown that the same batch of antiserum was not cytotoxic for bone marrow and that at higher concentrations it completely neutralised bone marrow colony formation stimulated by crude L cell-conditioned medium⁶.

Almost all the *in vitro* systems in which macrophage proliferation has been described have, as an obligatory requirement, the presence of either medium conditioned by certain cell lines^{7,17,18}, conditioned medium from cultures of primary cells⁷, embryo extract¹⁹ or inflammatory exudates^{20,21}. Many of these stimulatory sources (for example, L cell-conditioned medium, embryo extract and certain inflammatory exudates) have been shown to contain CSF^{6,22}. Our results clearly demonstrate that *in vitro* proliferation and differentiation within the mononuclear phagocytic line progression is dependent on the presence of a particular type of CSF. Mononuclear phagocytic cells reported to be stimulated to proliferate in culture include peritoneal macrophages^{8,20,21}, peritoneal exudate macrophages^{7,8}, alveolar macrophages^{17,18,13}, pleural exudate macrophages²³, lymph node and thymic cells²⁴, as well as blood monocytes¹⁴. To this list we now add Kupffer cells. We have shown that proliferation of mononuclear phagocytic cells from most of these sources is stimulated at 100 pM concentrations by a CSF sharing antigenic determinants with the circulating CSF⁶ present in normal mouse serum at concentrations of 150 pM (ref. 25). This suggests the existence of a common humoral mechanism for the stimulation of proliferation of mononuclear phagocytic cells irrespective of their tissue location. That this particular subclass of CSFs is a specific stimulator of mature macrophage production is further indicated by its inactivity on other cell types⁵, its action at extremely low concentrations^{3,5} and the fact that bovine serum albumin and transferrin are the only other macromolecular components required for CSF-stimulated bone marrow colony formation to occur²⁶ (L. J. Guilbert, personal communication). We feel that the criterion of pro-

Table 2 Neutralisation of mouse L cell-conditioned medium stimulated macrophage colony formation by antiserum to purified mouse L cell CSF

Source of CSF	Addition	Bone marrow (colonies per 5×10^4 cells)	Pulmonary alveolar cells	Blood mononuclear cells*	Non-parenchymal liver cells	Peritoneal exudate cells	Resident peritoneal cells
					Colonies per 10^3 cells		
None	None	0	0	0	0	0	0
L cell-conditioned medium	PBS	122	184	79	62	107	1
L cell-conditioned medium	Pre-immune serum	98	150	87	70	110	1
L cell-conditioned medium	Antiserum	39 (60%)	53 (65%)	25 (71%)	26 (63%)	37 (66%)	0

Pre-immune serum and antiserum were heat inactivated ($56^\circ\text{C}/30$ min) and diluted 1/16 with phosphate-buffered saline (PBS) before use. Mixtures of 2 parts L cell-conditioned medium to 1 part PBS or 1 part 1/16 pre-immune serum or 1 part 1/16 antiserum were incubated at room temperature for 60 min, before incubating each mixture with each cell population at 10% of the final culture volume. Figures represent the average colony count for duplicate cultures.

* Results expressed as colonies per 10^3 cells although there were 3×10^3 blood mononuclear cells per culture.

liferative responsiveness to this type of CSF will complement other criteria that have been applied²⁷ to delineate those cells of the reticuloendothelial system²⁸ that are of haemopoietic origin (the mononuclear phagocytic system) from those of mesenchymal origin.

Interestingly, the apparently more immature precursor cells in bone marrow require lower concentrations of CSF for optimal response than do the apparently more mature responding cells in lung, liver, peripheral blood and peritoneal exudates. The molecular basis of this decreasing sensitivity to CSF with maturation may be related to the different culture systems used (agar culture for bone marrow, liquid culture for the others), to production of inhibitory substances by the more mature cells of the series (for example, prostaglandin production²⁹), or to differences intrinsically related to CSF action.

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Autoimmune and polyclonal B cell responses during murine malaria

INTRACELLULAR protozoan infections of humans and rodents are often characterised by marked hypergamma-globulinaemia reflecting extensive activation of host B lymphocytes. In addition, the immunoglobulin (Ig) produced is directed not only against parasite antigens but also against a variety of unrelated antigenic determinants, and in some cases has been shown to contain antibodies reactive to certain host components (reviewed in refs 1 and 2). These findings have led to the suggestion that the infectious organisms may possess nonspecific or polyclonal activating properties³. Indeed, direct evidence for such mitogenic factors prepared from malarious mouse red blood cells (MRBC) has recently been obtained⁴. However, in most of these studies the use of serological or proliferative assays has provided only limited information on the different mechanisms whereby B cells are stimulated to produce antibody during the course of infection. I report here the use of plaque-forming cell (PFC) assays to reveal the patterns of both total and antigen-specific splenic B lymphocyte activation and also to define in more detail an anti-erythrocytic autoimmune response following malaria infection of mice. The results demonstrate that a specific, well-defined anti-erythrocytic response occurs shortly after infection which can be distinguished from, and induced independently of, a very large nonspecific polyclonal response. Both these responses are shown to be T-cell dependent.

BALB/c mice, 6–8 weeks of age, were injected intraperitoneally (i.p.) with 10^6 MRBC infected with either the lethal (17XL) or the nonlethal strain (17XNL) of *Plasmodium berghei* yoelii. Only results for lethal malaria are shown in this paper. Polyclonal B cell responses were measured using a modified PFC assay⁵ in which sheep red blood cells coated with a polyvalent sheep-anti-mouse immunoglobulin (α MIg-SRBC)

were used as indicator cells. This assay detects the production of PFC of all specificities and classes and is thus a measure of polyclonal activation. Plaque-forming cell responses specific for SRBC and the trinitrophenyl hapten (TNP) (ref. 6) were also measured. Autoimmune responses were assayed using bromelain-treated mouse red blood cells (BrMRBC) as indicators according to the technique of Cunningham⁷.

The results show that the total number of Ig-secreting cells in spleens (Fig. 1a) first increases above background at day 3 and rises rapidly until day 7 (the usual time of death in 17XL malaria). This represents a 100-fold increase in total splenic Ig-secreting cells. It is of interest that in self-limiting infections with 17XNL *P. berghei yoelii*, almost identical response kinetics of total PFC, peaking at day 7, are observed (not shown). As the parasitaemia in nonlethal infections is only 4% at 7 d compared with 70% in lethal infections, the number of Ig-secreting cells cannot depend on the level of parasitaemia.

When various antigen-specific responses are compared, two different patterns emerge. That of the anti-TNP PFC response (Fig. 1c), which peaks around day 7, is similar to that seen for total PFC; the anti-BrMRBC (Fig. 1d) and anti-SRBC (Fig. 1b) responses show distinct earlier peaks at day 4. These differing kinetics suggest that the observed responses result

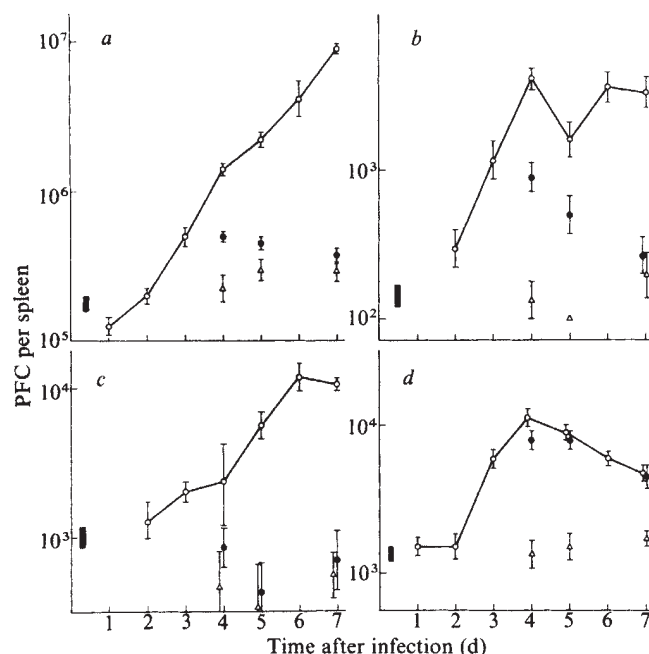


Fig. 1 Total and antigen-specific responses (○) in spleens of BALB/c mice (Cumberland View Farms, Clinton, Tennessee), plotted as log PFC against time following i.p. injection of 10^6 MRBC infected with *P. berghei yoelii*. a, Total Ig-secreting cells; b, direct anti-SRBC PFC; c, direct anti-TNP PFC; d, anti-BrMRBC PFC. Points represent the geometric means $\pm$ s.e.m. of data pooled from eight experiments in the case of a, b and d and three experiments in c. Note the differences in scales used. In four of the eight experiments (two experiments in c) aliquots of the 17XL *P. berghei yoelii*-infected MRBC were exposed to 20,000 R irradiation before injection (10^6 per mouse) and spleens were assayed for PFC at days 4, 5 and 7 thereafter (●). Points represent pooled data. In these mice, parasitaemias determined by Giemsa stains were always $<0.1\%$. In two control experiments, mice received equivalent numbers (2×10^6) of normal uninfected MRBC exposed to 20,000 R and their spleens were also assayed for PFC at days 4, 5 and 7 (△). Solid bars represent background levels of PFC from a pool of 20 or more mice expressed as a geometric mean $\pm$ s.e.m. All assays were done using a modified haemolytic PFC assay⁹ using normal SRBC, TNP-SRBC and BrMRBC as indicator cells. Total PFC were counted using SαMIg-SRBC (coupled with CrCl_3) as targets and a polyvalent rabbit-anti-mouse Ig as a developing serum.

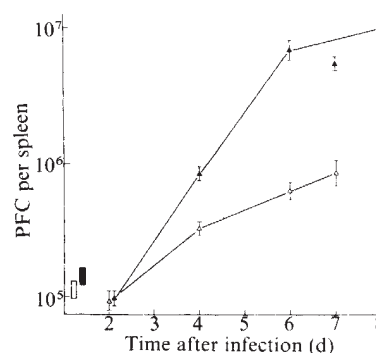


Fig. 2 Kinetics of the total splenic PFC response following infection of BALB/c nude mice (△) and their littermate controls (▲) (Sprague-Dawley) with 10^6 MRBC infected with 17XL *P. berghei yoelii*. Data are expressed as in Fig. 1. Points represent the pooled data of four experiments. Open (nude) and closed (littermate) bars represent the mean background levels obtained from six mice.

from two different mechanisms, one in which clones are directly stimulated by antigens present on or in parasitised cells (day 4 peaks) and one in which B cells are triggered nonspecifically by mitogenic factors (day 7 peaks).

To distinguish between these, advantage was taken of the fact that parasitised MRBC are rendered noninfectious following exposure to 20,000 R irradiation⁸. Thus, it was speculated that injection of 10^6 irradiated parasitised MRBC, which elicit no parasitaemia, would thereby eliminate any mitogenic effects caused by replicating parasites and reveal B cell clones activated specifically by the antigens on the parasitised erythrocytes. The results of such experiments are included in Fig. 1. Assays were done on days 4, 5 and 7. Compared to the 100-fold increase in total PFC seen following injection of infectious cells, only a 2–3-fold increase in Ig-secreting cells occurred when these cells were irradiated before injection. Similarly, no increase in anti-TNP PFC was observed in this situation. These findings suggest that most B cell triggering requires the nonspecific events which occur during infection. The possibility that massive specific stimulation by antigenically complex parasites accounts for the increase in total (or anti-TNP) PFC is unlikely, because reinfection of immune mice previously given 17XNL results in large increases in Ig-secreting cells in the absence of any parasitaemia (unpublished observation).

In contrast to the above, anti-BrMRBC responses were elevated in recipients of irradiated infected cells. These self-reactive PFC are thus unlikely to be nonspecifically induced, but are apparently stimulated directly by determinants on the infected erythrocytes which may be native to the parasite or modifications in the red cell membrane induced by the parasite. The pattern of the anti-SRBC response in mice given either normal or irradiated infected cells is of interest as the existence of both specific and nonspecific increases is evident. In recipients of non-infectious cells a smaller but still significant increase at day 4 was observed, but the remainder of the response (days 5–7) was completely ablated and may therefore be regarded as a result of nonspecific stimulation similar to that responsible for the anti-TNP response. Known cross-reactivity between BrMRBC and SRBC¹⁰ provides an explanation for the specific day 4 anti-SRBC response in mice injected with irradiated cells. It should also be noted (Fig. 1) that in no case does the injection of irradiated, normal uninfected mouse erythrocytes result in any significant PFC responses.

It is well documented that T cell proliferation occurs during malaria infections and that the course of the disease is different in T-cell deficient mice^{11–13}. To assess the role of T cells in the observed antibody-forming cell responses, adult athymic (nude) mice (5th backcross on a BALB/c background) and their littermates were infected i.p. with 10^6 17XL *P. berghei yoelii*. The results of such experiments (Fig. 2) demonstrate that

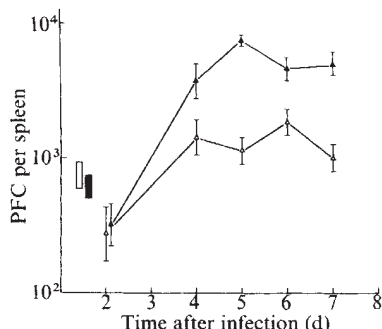


Fig. 3 Kinetics of the splenic direct PFC response to BrMRBC following infection of BALB/c nude mice (Δ) and their littermates ($\blacktriangle$) with 10^6 17XL *P. berghei yoelii*. Details are similar to those described in the legend to Fig. 2.

in nude mice the dramatic increase in Ig-secreting cells, always observed in their thymus-bearing littermates, does not occur; increases in nude mice usually range over 2–5-fold only, despite higher parasitaemias. Therefore, in malaria infections the observed polyclonal activation does not seem to result from direct stimulation of B lymphocytes by mitogenic factors. Instead, the data are more compatible with a mechanism in which mitogen-induced T cell proliferation causes the release of factors which, in turn, signal B cell differentiation into PFC. The findings of Weinbaum *et al.* that supernatants prepared from 17XL-infected MRBC induced predominantly T cell proliferation following culture with spleen cells¹⁴ agrees with this view. A lack of direct evidence for defective B cells in nude mice and the existence of normal background levels of splenic Ig-secreting cells in the nude mice used in these experiments (Fig. 2) argue against the possibility that the B cells in infected athymic mice lack the ability to be triggered.

Of particular importance in this study is the 10-fold increase in the number of PFC against autologous erythrocytes following injection of parasitised cells. Although varying numbers of anti-BrMRBC PFC occur when several strains of mice are compared, enhancement of normal levels has been achieved only indirectly by injection of either lipopolysaccharide or anti-lymphocyte serum⁷. In my experiments using irradiated infected cells, the anti-BrMRBC response could be seen in conditions which eliminate the possibility of nonspecific triggering, as shown by the lack of an anti-TNP PFC response (Fig. 1c,d). Experiments with nude mice (Fig. 3) further showed that this increase in BrMRBC PFC, which is totally of the IgM class, is a T-cell dependent process. *Babesia* infections of nude mice¹⁵ have also indicated a T-cell dependent anti-BrMRBC response.

Based on these findings, the increased auto-reactivity following infection with lethal malaria apparently results from the generation of helper T cells specific for membrane-associated determinants on parasitised mouse erythrocytes. These then collaborate with the B cell population recognising the antigens exposed by bromelain treatment. The antigens in question may not be superficial, as PFC assays using parasitised MRBC as targets have not as yet revealed PFC. Recent experiments have further shown that the kinetics of an anti-BrMRBC response can be used to distinguish between lethal and nonlethal malaria infections (work in preparation).

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Oestrogen content of the embryonic rabbit ovary

ALTHOUGH the essential role of endocrine function of the fetal testis for male phenotypic development is established^{1–3}, endocrine function of the fetal ovary is not thought to be essential for development of the female phenotype¹. Furthermore, it has been generally assumed that the fetal ovary has little capacity for oestrogen synthesis until late in embryonic life^{4–9}. However, we recently reported that the capacity of the embryonic ovary of rabbit and human to synthesise oestrogen from radioactive androgen develops at about the same time as the onset of the capacity for testosterone synthesis in the fetal testes of the same species and before the histological differentiation of the ovary^{10,11}. To determine if this enzymatic capacity to synthesise oestrogens is actually indicative of oestrogen formation in the intact tissue, we have measured and report here the content of 17β -oestradiol and testosterone in embryonic gonads of the fetal rabbit at varying stages of development. The results indicate that the onset of formation of the characteristic steroid hormones takes place simultaneously in ovary and testis.

Gonads were pooled from embryos that varied from day 17 to day 28 of development. This period of development was chosen because it encompasses the period between the end of the indifferent phase of sexual development (day 17) and birth (usually day 30 or 31). Histological differentiation of the Leydig cell is well advanced by day 19 (ref. 12), and male phenotypic differentiation is mainly finished by day 22 (ref. 1). In contrast, histological differentiation of the fetal ovary has not been recognised until late in embryonic development¹. At the earliest stage (day 17) 50–70 gonads were combined, and no attempt was made to separate the sexes; at subsequent stages 10–40 gonads from each sex were combined separately for each pool. 17β -Oestradiol content was low at day 17, rose slightly in ovaries at day 18, and by day 19 was clearly higher than in testes (2.2 compared with 0.6 pg per gonad) (Fig. 1). In contrast, the testosterone content of the testis was higher than that of the ovary at each stage examined from day 18 onwards. When these data are expressed in terms of hormone concentration (amount per mg protein) the differences are even more striking (Fig. 2). From day 18 onwards the concentration of 17β -oestradiol is higher in ovary than in testis and peaks at a level 20-fold higher than the corresponding level in testes at day 19 of gestation. Although the concentration of testosterone was always greater in the fetal testis than in the fetal ovary, the concentration of testosterone in the ovary on day 19 (about 300 pg per mg protein) is sufficient to serve as substrate for the formation of 17β -oestradiol (30 pg per mg protein). No significant differences were seen in the concentration of 17β -oestradiol in the two sexes between days 18 and 28 of development in plasma ($53.7 \pm \text{s.e.m. } 16.1 \text{ pg ml}^{-1}$ in females and $77.5 \pm \text{s.e.m. } 25.1 \text{ pg ml}^{-1}$ in males) or brain ($0.4 \pm 0.2 \text{ pg per mg protein}$ in females and $1.6 \pm 0.8 \text{ pg per mg protein}$ in males).

Thus, the increase in the content and concentration of 17β -oestradiol in the fetal ovary is identical to the developmental

pattern of the enzymes necessary for oestrogen synthesis from radioactive androgen in the fetal rabbit ovary¹⁰. The finding that the content of 17β -oestradiol is at all times higher in ovary than in testis and that the increase in the content of 17β -oestradiol in ovary apparently occurs at the same time as the onset of testosterone synthesis and also the accumulation of testosterone in the fetal testis indicates that the onset of the characteristic endocrine function occurs at the same time in the gonads of the two sexes. This does not mean that other aspects of gonadal differentiation may not occur at different times in ovary and testis; indeed, organisation of germ cells may occur before day 18 (ref. 1). However, the finding that the onset of oestrogen synthesis by the fetal ovary occurs simultaneously with the onset of testosterone synthesis by the testis indicates that ovarian differentiation takes place earlier than has been recognised previously and that further studies of gonadal differentiation will have to take this finding into account. Whether early oestrogen synthesis in ovary has functional significance or whether it is simply a cellular marker of differentiation is not clear. Because elevated concentrations of plasma 17β -oestradiol were not observed in the female

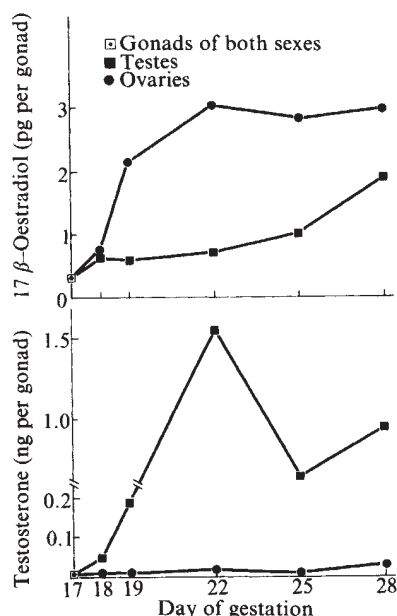


Fig. 1 Oestrogen and testosterone content of fetal rabbit gonads. Gonads were dissected from fetal rabbits at the times indicated and stored frozen. After a complete series had been collected, approximately 3,000 d.p.m. each of $[1,2,6,7-^3\text{H}]$ testosterone (85 Ci mmol^{-1}) and $[2,4,6,7,16,17-^3\text{H}]17\beta$ -oestradiol (151 Ci mmol^{-1}) corresponding to 5 and 2.5 pg, respectively, were added to each sample as recovery standards. The samples were then extracted with 5 ml chloroform:methanol (2:1, vol:vol). The chloroform layer was backwashed twice with 1 ml water, taken to dryness under a stream of nitrogen and reconstituted in 5 ml of 2% ethyl acetate in iso-octane. Testosterone and 17β -oestradiol were then separated by celite:ethylene glycol-water column partition chromatography as described¹¹. The average recoveries were greater than 85% both for testosterone and for 17β -oestradiol. Testosterone was measured by radioimmunoassay with a limit of sensitivity of 5 pg (ref. 14). The inter- and intra-assay coefficients of variation were 8 and 4%, respectively. 17β -Oestradiol was measured by radioimmunoassay (Micomedic) using two aliquots corresponding to 20 and 40% of the total column eluate, with a limit of sensitivity of 1 pg. With the larger aliquot, values for 17β -oestradiol were between 10 and 25 pg in fetal ovaries. Total 17β -oestradiol in the no-tissue blank was 1.7 pg. The inter- and intra-assay coefficients of variation for the 17β -oestradiol assay were 8 and 3%, respectively. Both assays were linear with increasing aliquots of tissue samples. The data are expressed as the mean of three separate experiments for 17β -oestradiol and the mean of two experiments for testosterone.

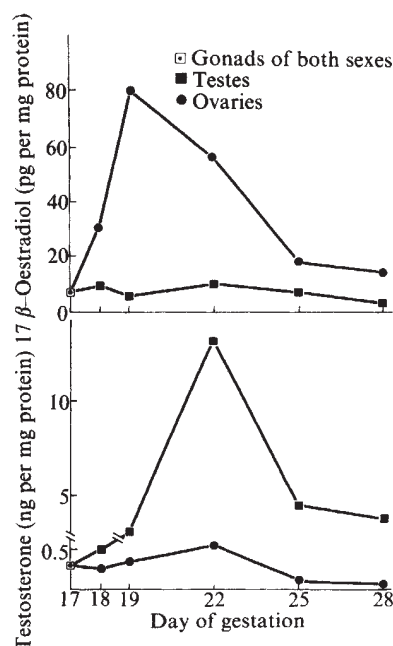


Fig. 2 17β -Oestradiol and testosterone concentration of fetal rabbit gonads. The data described in Fig. 1 have been expressed as the amount of each hormone recovered per mg tissue protein. Tissue protein was determined as described¹⁰ using the method of Lowry *et al.*¹⁵ with bovine serum albumin as the standard. Data are expressed as the mean of three experiments for 17β -oestradiol and the mean of two experiments for testosterone.

embryo and as endocrine function of the embryonic ovary is not essential for the formation of the female phenotype, any effect of ovarian 17β -oestradiol would probably be exerted locally within the developing ovary. Indeed, as oestrogen has the capacity to induce follicular development in the immature ovary¹³ and as the onset of oestrogen synthesis precedes histological differentiation of the primordial follicles, it is possible that locally synthesised oestrogen functions to initiate ovarian growth and development.

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Inhibition of prolactin secretion by histidyl-proline-diketopiperazine

ON the basis of its activity as a thyrotropin-releasing factor, the tripeptide pyroGlu-His-Pro-NH₂ (TRH) has been isolated from the hypothalamus^{1,2}. Subsequently, the tripeptide was found in extrahypothalamic tissues of vertebrates and invertebrates and its activity as a prolactin-releasing factor and as a neurotropic has been recognised (for review see ref. 3). However, the way in which these different activities are elicited at different sites is still unknown. According to the concept of limited proteolysis⁴, distinct functions at different sites could emerge if TRH were susceptible to specific enzymatic attack. Degrading enzymes might therefore not only be responsible for the regulation of TRH concentration⁵⁻⁷, which in turn controls both the degree and the duration of the stimulatory effects of the tripeptide, but could also give rise to new activities. We report here the degradation of TRH by enzymes in serum, hypothalamus, pituitary and brain and the identification of the degradation products. We have also studied a presumptive metabolite, histidyl-proline-diketopiperazine, which seems to inhibit prolactin release.

Although the rapid inactivation and metabolism of TRH has been reported by several investigators (for review see ref. 8) so far the mechanisms underlying the fragmentation have not been fully elucidated. To investigate the enzymatic degradation of TRH, [³H-His]TRH was incubated with a number of enzyme preparations. The radiolabelled products were separated by thin layer chromatography and identified by cochromatography with synthetic compounds (Fig. 1).

After incubation of [³H-His]TRH with serum, only His-Pro-NH₂ was identified as the primary cleavage product. The enzyme responsible is an apparently TRH-specific pyrrolidone-carboxyl-peptidase of approximately 280,000 molecular weight (P. Nowak and K.B., unpublished). His-Pro-NH₂ is subsequently deamidated and then hydrolysed to the constituent amino acids. The primary product is converted during chromatography to histidyl-proline-diketopiperazine, as can be seen from the raised background between the positions of the two compounds (Fig. 1). This intramolecular condensation is a characteristic reaction of proline-containing dipeptide derivatives⁹. Incubation of [³H-His]TRH with extracts from pituitary, hypothalamus and brain yielded both histidyl-proline amide, which gives rise to histidyl-proline-diketopiperazine, and deamido-TRH as primary cleavage products (Fig. 1). Histidyl-proline was found as the only secondary intermediate before the free amino acids. The pyrrolidone-carboxyl-peptidase from these tissues (molecular weight ≈ 30,000), which unlike the serum enzyme is not specific for TRH but also splits LHRH

and synthetic substrates, and the TRH-deamidating enzyme (molecular weight ≈ 76,000) have been separated by conventional methods.

Attempts to demonstrate enzymatic formation of histidyl-proline-diketopiperazine have not been successful because of the rapid spontaneous conversion from histidyl-proline amide under the experimental conditions used. However, the structural features of the compound (Fig. 2), which like centrally acting drugs contains a lactam ring, prompted us to search for possible biological activity.

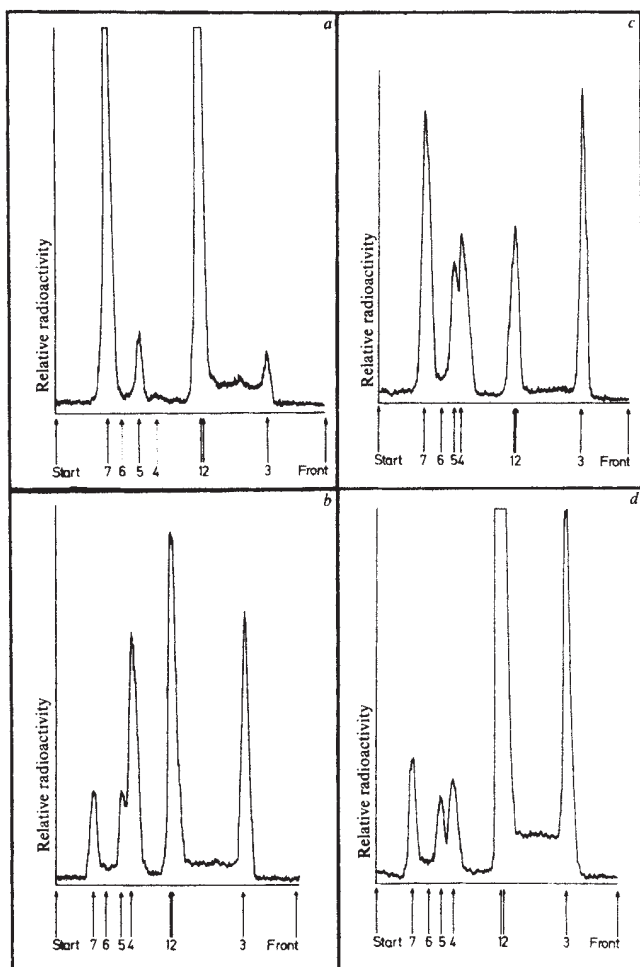


Fig. 1 Male Sprague Dawley rats (200 g body weight) were decapitated. Trunk blood was collected and after 3 h at 4°C serum was obtained by centrifugation at 2,000g for 15 min. Tissue immediately removed after decapitation was homogenised by a Teflon homogeniser in potassium phosphate buffer (100 mM, pH 7.4), containing 2 mM DTT, 0.5 mM EDTA. 30 volumes buffer for the homogenisation of the pituitary and hypothalamic tissue and 20 volumes for the brain tissue were used. After centrifugation at 27,000 g for 30 min the supernatant was aspirated. 1 μ Ci [³H-His]TRF (60 Ci mmol⁻¹), prepurified by thin layer chromatography as described below, was incubated for 30 min at 37°C with 20 μ l serum (diluted 1:3 with 100 mM Tris-HCl buffer, pH 7.4) or with 20 μ l of the tissue extract. The incubation was terminated by addition of 50 μ l methanol. After centrifugation, the supernatant, synthetic amino acids and peptides were applied to precoated thin layer plates (Merck 5721) and developed in vapour saturated tanks using the solvent system CHCl₃:CH₃OH:NH₃ aq. 125:75:25 (v/v). The radiolabelled split products were localised by scanning for radioactivity or by autoradiography and the cochromatographed standard material by staining with ninhydrin followed by Pauly reagent. 1, TRF; 2, His-Pro-NH₂; 3, His-Pro-diketopiperazine; 4, deamido-TRF; 5, His-Pro-OH; 6, pyroGlu-His-OH; 7, His. Enzyme source: a, serum; b, hypothalamus; c, brain; d, pituitary.

Table 1 Inhibition of prolactin secretion of GH₃ cells by histidyl-proline-diketopiperazine

Treatment	Dose (ng ml ⁻¹)	Prolactin (ng) per mg cell protein	
		Experiment 1 GH ₃ 1350	Experiment 2 GH ₃ 1382
Control	—	470 ± 67	970 ± 130
[His-Pro]	0.1	391 ± 67	862 ± 147
	0.5	283 ± 40*	865 ± 156
	1.0	227 ± 82*	762 ± 94*
	4.0	247 ± 34†	627 ± 40‡

GH₃ cells were cultured in synthetic medium containing heat-inactivated serum¹⁰. Thirty minutes after addition of the test substances to fresh medium, the incubation fluid was removed and assayed for prolactin using the NIAMD radioimmunoassay kit. Cell protein was determined by Folin's assay using BSA as standard.

* $P < 0.05$, † $P < 0.02$, ‡ $P < 0.01$ (Student's *t* test).

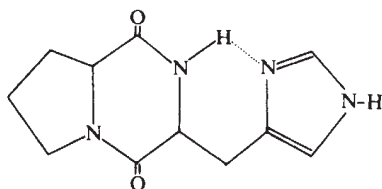


Fig. 2 Structure of histidyl-proline-diketopiperazine. 100 mg His-Pro-NH₂ was dissolved in water, the pH adjusted to 7.5 by the addition of 2 N NH₃ aq. and allowed to stand for 24 h. After evaporation *in vacuo*, the residue was dissolved in 0.5 ml methanol and the solution applied to precoated preparative silica gel thin layer plates (Merck 5745), which were developed using the solvent system CHCl₃:CH₃OH:38% acetic acid (90:67.5:30). The zone with *R_f* 0.42–0.50, containing the histidyl-proline-diketopiperazine, was scraped off and eluted with methanol. After evaporation the residue was dissolved in 100 mM acetic acid and stored at –80 °C. Magnetic resonance at 220 MHz showed chemical shifts at 7.5 (Im-C₂H), 6.8 (Im-C₄H), 4.5 (His-C^αH), 4.2 (Pro-C^αH), 3.2 (Pro-C^βH₂), 2.13 (His C^βH₂); 1.8 (Pro C^βH, C^γH₂). Mass spectroscopy at 155 °C showed dominant ions at *m/e* 234 (M+), 154 (proline-glycine diketopiperazine), 137, 122, 109 (histidine fragments), 82 and 81 (methyl-imidazole-ion), 70 (pyrroline ion).

We studied the effects of histidyl-proline-diketopiperazine on prolactin release in two experimental systems. Cells of the GH₃ tumour line, cultured in synthetic medium containing heat-inactivated serum, were incubated with increasing concentrations of the compound and the prolactin secreted into the medium within 30 min was determined by radioimmunoassay (Table 1).

While TRH stimulates the secretion of prolactin by the GH₃ cells, histidyl-proline-diketopiperazine decreased the release of prolactin in a dose-related manner to approximately 50% of the control levels. Further studies are necessary to determine whether an effective concentration of histidyl-proline-diketopiperazine can be formed directly by these cells, by a slow metabolism of TRH.

In a second type of experiment, trained proestrous rats were anaesthetised with urethane and the test substances administered 2 h later. One microgram of histidyl-proline-diketopiperazine significantly depressed the TRH-stimulated prolactin release (Table 2).

The degree of inhibition of prolactin release in both experimental systems is comparable to that observed elsewhere in the course of attempts to isolate a prolactin release inhibiting factor (PIF)¹¹. It is also noteworthy that the physicochemical characteristics described by Dhariwal *et al.*¹² for PIF, that is, a

slight basic charge, low molecular weight, prolonged retention by Sephadex G-25 and relative lability, are shared by histidyl-proline-diketopiperazine. These similarities might be purely accidental and therefore warrant further investigation.

We propose as a working hypothesis that enzymatic degradation could give rise to an inhibitory compound, which counterbalances the releasing effect of administered TRH and thereby limits the extent of prolactin secretion. Much work remains to elucidate the mechanisms mediating the action of histidyl-proline-diketopiperazine and to study the regulatory mechanisms which control the formation of the compound.

The recently described antagonistic effect¹³ of histidyl-proline-diketopiperazine on ethanol narcosis, which is more pronounced than that seen after TRH administration, is another example of the biological effects of the cyclised degradation product. In this case, however, histidyl-proline-diketopiperazine is the stimulatory rather than the inhibitory metabolite of TRH.

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Table 2 Inhibition of prolactin secretion of proestrous rats by histidyl-proline-diketopiperazine

Group	Treatment	Dose	Serum prolactin (ng ml ⁻¹)
1	NaCl	—	79.3 ± 17.7
2	TRF	1 µg	223.0 ± 38.0*
3	TRF + [His-Pro]	1 µg + 1 µg	132.0 ± 19.1†
4	[His-Pro]	1 µg	49.6 ± 11.0‡c

Sprague Dawley rats (180–200 g body weight) undergoing regular cycles (confirmed by taking vaginal smears for two recurrent cycles) were used. On the day of proestrous between 1000 and 1100 h the rats were anaesthetised by s.c. injection of a urethane solution (150 mg per 100 g body weight) and 2 h later the test substances, dissolved in 1 ml saline were injected i.v. within 1 min. Six minutes later the animals (eight rats for each group) were decapitated and the serum sampled. Prolactin was assayed using the NIAMD assay kit. Statistical evaluation (Student's *t* test): * *P* < 0.05, group 2 versus groups 1, 3 and 4; † *P* < 0.05, group 3 versus groups 2 and 4; ‡ *P* < 0.05, group 4 versus groups 2 and 3.

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Tricyclic antidepressants block histamine H₁ receptors of mouse neuroblastoma cells

TRICYCLIC antidepressants are structurally related to antihistamines and are known to possess antihistaminic properties¹. Green and co-workers^{2,24} showed that these drugs competitively antagonised histamine H₂ receptor-mediated cyclic AMP formation by homogenates of guinea pig brain. Using muscarinic acetylcholine receptor-mediated cyclic GMP formation by mouse neuroblastoma cells (clone N1E-115)³, the interactions of psychotherapeutic drugs with this cholinergic receptor were studied in this laboratory^{4,5}. While investigating the desensitisation of this muscarinic receptor⁶, it was found that histamine also stimulated cyclic GMP formation by activation of histamine H₁ receptors of these cells⁷. This assay for histamine H₁ receptors was used to obtain quantitative data for tricyclic antidepressant interactions with these receptors. This report shows that these drugs are potent competitive inhibitors of histamine H₁ receptors and that some of these compounds are amongst the most potent H₁ antagonists known.

Cyclic GMP formation by intact mouse neuroblastoma cells was determined by an assay involving radioactively labelling intracellular stores of GTP, the precursor of cyclic GMP, and isolating radioactively labelled cyclic GMP. Although it does not measure absolute levels of cyclic GMP, this assay yields results which are otherwise comparable to those obtained with radioimmunoassay⁸.

Dose-response curves for histamine were shifted to the right in the presence of all the tricyclic antidepressants tested (as for example, by amitriptyline, Fig. 1); additionally, at high agonist concentrations, the effects of the antagonist were overcome. Such results indicate competitive antagonism and allow the determination of the apparent equilibrium dissociation constant (K_B) for the antagonist-receptor complex from the equation⁹

$$K_B = \frac{[B]}{DR - 1}$$

where $[B]$ = concentration of antagonist and DR = dose ratio. K_B values which were determined in this manner for six tricyclic antidepressants ranged from 1×10^{-10} to 2×10^{-7} M (Table 1). Amitriptyline, the most potent histamine H₁ receptor antagonist of our series, was in other series the most potent antagonist of the histamine H₂ receptor² and the muscarinic acetylcholine receptor^{5,10,11}. The rank order of potency of these drugs at histamine H₁ receptor blockade was essentially the same as that for muscarinic receptor blockade in these mouse neuroblastoma cells (Table 1). Thus, compounds with tertiary amine side chains were more potent than those with secondary amine moieties.

Because amitriptyline was much more potent at blocking the histamine H₁ receptor in these cells than the H₁-selective antagonist pyrilamine ($K_B = 2 \times 10^{-9}$ M with these cells⁷, a value similar to that found for the drug in binding assays using guinea pig ileum¹²), its K_B value was determined many times. With $n = 12$ (that is, 12 experiments of which Fig. 1 represents one) the amitriptyline value ($\pm$ s.e.m.) was $1.3 \pm 0.3 \times 10^{-10}$ M. For comparison, the K_B for diphenhydramine, a commonly used antihistamine, was 2×10^{-8} M (data not shown), a value close to that found for this compound in studies of histamine induced contractions of guinea pig ileum¹³.

The K_B values for pyrilamine and diphenhydramine reported here are in reasonable agreement with those derived from studies with guinea pig ileum^{12,13}. Thus, in one respect the histamine H₁ receptors of clone N1E-115 are similar to those found in guinea pig ileum. This supports the idea that tricyclic antidepressants are in general H₁ receptor blockers. However,

histamine H₁ receptors of brain have not yet been characterised, and whether H₁ receptors of mouse neuroblastoma cells are similar to those of normal neurones *in vivo* is not yet known.

Green and Maayani² compared anti-H₂ potency with anticholinergic potency for three tricyclic antidepressants by comparing their data with those from the literature for these drugs and the muscarinic acetylcholine receptor^{10,11}. This comparison showed that the anti-H₂ potency of these compounds was very similar to their anticholinergic potency. A similar comparison for the six compounds of our series using data derived in this laboratory with these cells (Table 1) shows that the tricyclic antidepressants were always more potent at histamine H₁ receptor blockade than at muscarinic receptor blockade. (The anticholinergic potencies of these compounds in mouse neuroblastoma cells are in closer agreement with those of Rehavi *et al.*¹¹ using mouse brain than those of Snyder and Yamamura¹⁰ using rat brain). In addition, a comparison of new anti-H₁ data for amitriptyline and imipramine with the published anti-H₂ data² for these drugs shows that they are very much more potent at blocking H₁ than H₂ receptors (Table 1). Finally, all of the compounds listed in Table 1 except desipramine apparently have a much higher affinity for histamine H₁ receptors than for α -adrenergic receptors¹⁴ which are antagonised better by tertiary amine tricyclic antidepressants than by their secondary amine analogues (Table 1).

U'Pritchard *et al.*¹⁴ showed that the apparent affinities of tricyclic antidepressants for central α -adrenergic receptors correlated well with their abilities to cause sedation and hypotension. Since the rank order for tricyclic antidepressants at α -receptor blockade is nearly the same as that for H₁-receptor blockade (Table 1), it follows that the antihistaminic properties of these drugs also correlate well with their abilities to cause

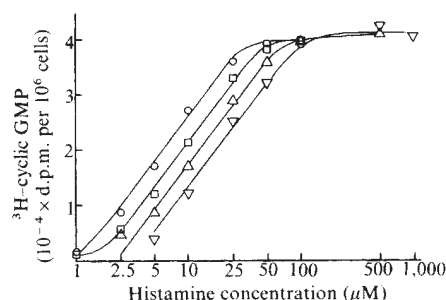


Fig. 1 Effect of a tricyclic antidepressant drug on the histamine dose-response curve. Mouse neuroblastoma clone N1E-115 cells (subculture 16) were cultured and assayed for histamine-stimulated cyclic GMP formation as described previously^{4,5} except for the following modifications (described in detail elsewhere⁸): (1) [³H]guanine (1 μ M final concentration, 10 μ Ci ml⁻¹) was used to label intracellular stores of GTP; (2) HEPES buffer and the phosphodiesterase inhibitor were not used; and (3) ³H-cyclic GMP from the cation exchange resin was further fractionated by precipitation with ZnCO₃ (ref. 21) to remove some contaminants (mostly ³H-GDP and ³H-GTP)²². Thus, 25 μ l each of 3.2M ZnSO₄ and 3.2M Na₂CO₃ were added to the 1.5 ml fraction from the cation ion exchange column. After sedimentation of the precipitate, the supernatant was quantitatively removed and counted for radioactivity as before^{4,5}. Cells were incubated with amitriptyline (Merck Sharp and Dohme) for 30 min before addition of histamine dihydrochloride (Sigma) for 30 s at the indicated concentrations. Each point was determined in duplicate. Basal radioactivity (no histamine) which was subtracted from these data averaged ($\pm$ s.e.m.) 1,700 $\pm$ 200 d.p.m. per 10⁶ cells. Maximum stimulation by histamine above basal was therefore about 23-fold. There were approximately 2.2×10^5 cells and 0.5 mg protein per assay. Cell counts were determined with a Coulter counter (model Z₁) and proteins were determined by a modification of the method of Lowry *et al.*²³ with serum albumin as standard. $\circ$, Control; $\square$, 5×10^{-11} M amitriptyline added; $\triangle$, 1×10^{-10} M amitriptyline; ∇ , 2×10^{-10} M amitriptyline.

Table 1 Tricyclic antidepressant antagonism of histamine, muscarinic acetylcholine and α -adrenergic receptors

Compound	K_B for H_1 receptor*	K_B for H_2 receptor†	K_B for muscarinic receptor‡	K_i for α -adrenergic receptor§
Tertiary amines				
Amitriptyline	0.1	45	100	24
Doxepin	2	160	300	23
Imipramine	5	250	400	58
Secondary amines				
Nortriptyline	6	790	1,000	71
Protriptyline	30	2,000	2,000	277
Desipramine	200	1,300	2,000	148

* Using mouse neuroblastoma cells, each compound was tested at two or three different concentrations in a manner similar to that described in Fig. 1. Apparent K_B values were determined as described in the text. Average values are reported, in nM.

† Guinea pig brain (from ref. 24).

‡ Mouse neuroblastoma cells (from ref. 5).

§ Rat brain (from ref. 14).

sedation and hypotension. Because sedation is the most common side effect of H_1 receptor antagonists, they are used clinically as sedative/hypnotics¹⁵⁻¹⁷. There is no evidence to suggest that these antihistamines have antidepressant effects or that they also block α receptors. The higher apparent affinities of some tricyclic compounds for H_1 receptors than for α receptors suggests that *in vivo* their sedative effects may be mediated by H_1 receptor blockade. The hypotensive effects of these drugs cannot be ascribed to blockade of central α receptors since such a receptor blockade would be expected to cause an increase rather than a decrease in blood pressure¹⁸. In addition, there is evidence to suggest that H_1 receptor antagonism causes hypotension whereas H_2 receptor antagonism causes hypertension¹⁹. The latter effect on blood pressure may involve central H_2 receptors²⁰.

What is most clear, however, is that tricyclic antidepressants are antagonists of histamine H_1 , histamine H_2 , muscarinic acetylcholine and α -adrenergic receptors as well as biogenic amine transport systems. Which, if any, of these biochemical effects causally relates to their various clinical effects remains to be determined.

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The nature of an initiator constitutive mutation in *Aspergillus nidulans*

ONLY a few *cis*-acting regulatory mutations have been described in eukaryotes. One of these, *uap-100*, affects the expression of *uapA*, the adjacent putative structural gene for a uric acid-xanthine permease in the ascomycete *Aspergillus nidulans*¹. Only one protein coded by a formally defined regulatory gene has even been tentatively identified from an eukaryote. This is the positively acting product of the *uaY* gene of *A. nidulans* (D. Philippides and C.S., in preparation). As the *uapA* gene is under the control of *uaY* (ref. 2 and this paper) the interactions between *uap-100* and wild type and mutant *uaY* products are of considerable interest. Here we show that the constitutivity conferred by *uap-100* is strictly dependent on a functional *uaY* product. This is compelling evidence for the direct involvement of the *uaY* product in the regulation of *uapA* and, by implication, of other genes under *uaY* control, and might indicate that the *uaY* product exists as at least two conformers in equilibrium.

The uric acid-xanthine permease is subject both to induction by uric acid and some of its analogues¹ and to repression by ammonium^{1,3}. Presumably, therefore, the *uapA* gene must be controlled by an adjacent region comprising receptor sites for the positively acting products of the pathway-specific *uaY* gene (ref. 4 and D. Philippides and C.S., in preparation), which mediates induction, and the more general *areA* gene³ which mediates ammonium repression. The *uap-100* mutation exerts at least three pleiotropic effects: (1) a 2.5-fold 'up-promoter' effect (that is, the fully induced level of *uapA* permease is 2.5 times greater than in the wild type); (2) initiator constitutivity such that the uninduced level of uric acid transport activity is more than 62% of the fully induced level (whereas in the wild type the uninduced level is less than 16% of the induced level); (3) the ability to accommodate the *areA* product resulting from the *areA-102* mutation (whereas this mutant *areA* product cannot elicit the *uapA* expression in strains wild type for the *cis*-acting regulatory region for *uapA*). It is worth emphasising, however, that although *uap-100* alters an *areA* product binding site, the available evidence indicates that it does not alleviate the stringent requirement for the *areA* product¹.

As *uap-100* results in initiator constitutivity, largely bypassing the need for induction by effectors of the *uaY* product such as 2-thiouric acid^{1,5}, it would be expected that *uap-100* would also alleviate the requirement for the *uaY* product. This would result in suppression of *uaY*⁻ mutations by *uap-100*. It is possible to inquire whether the *uap-100* mutation suppresses several different manifestations of the permease-less phenotype of *uaY*⁻ strains. For example, one can ask whether *uap-100* restores the uptake of labelled uric acid. Alternatively, one can take advantage of the fact that 2-thiouric acid, a substrate of the *uapA* permease, inhibits the laccase-catalysed formation of green conidial pigment^{1,2,6,7}. *uaY*⁻ strains form green pigment even in the presence of 2-thiouric acid, presumably

Table 1 Uric acid uptake by wild type and mutant strains

Relevant genotype	Uninduced	¹⁴ C/ ³ H Induced
Wild type	0.0481	0.1643
<i>uap-100</i>	0.2891	0.6091
<i>uaY-9</i>	0.0361	0.0243
<i>uaY-9 uap-100</i>	0.0155	0.0193

The method used for uptake studies and its rationale have been described previously^{1,14}. Uric acid uptake is measured by ¹⁴C counts, and ³H counts measure protein synthesis and hence growth. In addition to the *uap* and *uaY* markers specified above, all strains carry *yA-2* (yellow conidial colour), *luA-1* (L-leucine auxotrophy) *hxB-13* (loss of xanthine dehydrogenase), and *uaZ-11* (loss of urate oxidase). Mycelia were grown for 21 h at 37°C on solid glucose-containing minimal medium¹⁵ supplemented with (final concentrations) 10 µg l⁻¹ biotin, 5 mM urea, and 1 mM 4,5-³H-L-leucine at a final specific activity of 500 mCi mol⁻¹ either without (uninduced) or with (induced) 27.7 µM 2-thiouric acid. Mycelia were incubated for 30 min at 37°C in uptake medium, liquid glucose minimal medium¹⁵ supplemented with (final concentrations) 10 µg l⁻¹ biotin, 5 mM urea, 1 mM L-leucine (unlabelled) and 59.5 µM 2-¹⁴C-uric acid at a final specific activity of 3.36 Ci mol⁻¹.

because this analogue is not transported into the cells. Thus, suppression of *uaY*⁻ mutations by *uap-100* would result in a double mutant class, which although still lacking xanthine dehydrogenase and urate oxidase⁸ and therefore still unable to grow on hypoxanthine and uric acid as nitrogen sources, would be sensitive to 2-thiouric acid. This prediction proved false for each of two *uaY*⁻ alleles: *uaY-9*, a nonleaky point mutation

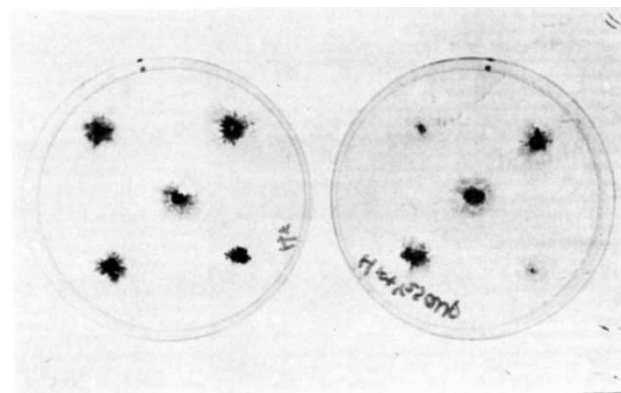


Fig. 1 Inhibition of growth of heterokaryons by 220 nM allopurinol on hypoxanthine as nitrogen source. An example is shown of the requirement for a *uaY*⁺ allele to be in the same nucleus as the *uap-100* mutation for the latter to be expressed. The relevant heterozygous controls are also shown. Plates: left, 700 µM hypoxanthine as nitrogen source; right, 700 µM hypoxanthine as nitrogen source plus 220 nM allopurinol. Heterokaryons: centre *uaY*⁺ *uap*⁺/*uaY*⁺ *uap*⁺; top left *uaY*⁺ *uap*⁺/*uaY*⁺ *uap-100*; top right *uaY*⁺ *uap*⁺/*uaY-9* *uap*⁺; bottom left *uaY-9* *uap-100*/*uaY*⁺ *uap*⁺; bottom right *uaY-9* *uap*⁺/*uaY*⁺ *uap-100*. These heterokaryons correspond respectively to nos 1, 2, 4, 6 and 7 in Table 2.

and *uaY-207*, a putative short deletion mutation (ref. 8 and N. Sdrin and C.S., in preparation). In both cases *uaY*⁻ *uap-100* double mutants were indistinguishable from *uaY*⁻ single mutants and could only be identified by progeny testing.

The failure of *uap-100* to suppress *uaY*⁻ mutations for 2-thiouric acid sensitivity can be explained in two ways: (1) in addition to uptake there is an as yet unidentified intracellular activity under *uaY* control which is necessary for 2-thiourate toxicity; (2) the *uap-100* mutation does not bypass the need for the *uaY* product for *uapA* expression. Table 1 shows that the second explanation is correct: an *uaY-9* *uap-100* double mutant is as defective in uric acid uptake as an *uaY-9* single mutant. This experiment does not exclude the possibility that the first mechanism might also be operative.

The activity of the *uaY* gene as tested in heterokaryons is apparently limited to the nucleus^{9,10}. This might arise from a dose effect resulting from the limiting concentration of *uaY*

Table 2 Allopurinol sensitivity of heterokaryons and diploids

Relevant genotype of heterokaryon or diploid	Growth on hypoxanthine as nitrogen source		Growth on hypoxanthine as nitrogen source in the presence of 220 nM allopurinol	
	Heterokaryon	Diploid	Heterokaryon	Diploid
(1) <i>uaY</i> ⁺ <i>uap</i> ⁺ / <i>uaY</i> ⁺ <i>uap</i> ⁺	+	+	+	+
(2) <i>uaY</i> ⁺ <i>uap</i> ⁺ / <i>uaY</i> ⁺ <i>uap-100</i>	+	+	-	-
(3) <i>uaY</i> ⁺ <i>uap-100</i> / <i>uaY</i> ⁺ <i>uap-100</i>	+	+	-	-
(4) <i>uaY-9</i> <i>uap</i> ⁺ / <i>uaY</i> ⁺ <i>uap</i> ⁺	+	+	+	+
(5) <i>uaY-9</i> <i>uap</i> ⁺ / <i>uaY-9</i> <i>uap</i> ⁺	-	-	-	-
(6) <i>uaY-9</i> <i>uap</i> ⁺ / <i>uaY</i> ⁺ <i>uap-100</i>	+	+	-	-
(7) <i>uaY-9</i> <i>uap-100</i> / <i>uaY</i> ⁺ <i>uap</i> ⁺	+	+	+	-

Heterokaryons were established on 10 mM NaNO₃ as sole nitrogen source, following the procedures routinely used in *A. nidulans*¹⁶. The following markers were used: *yA-2* (yellow conidiospores), *waA-4* (white conidiospores), *fwA-1* (fawn conidiospores) *biA-1* (requirement for biotin), *pabaA-1* (requirement for para-aminobenzoic acid) *pyroA-4* (requirement for pyridoxine) *pantoB-100* (requirement for pantothenic acid). Equilibrated heterokaryons (with the exception of no. 5) were pregrown on 700 µM hypoxanthine before transferring to the testing media. Hypoxanthine in the testing media was also added to a final concentration of 700 µM. Diploids were isolated following the routine procedures in use in *A. nidulans*. Homozygous wild type (no. 1), *uap-100* (no. 3) and *uaY*⁻ (no. 5) diploids and heterokaryons were included in addition to the proper heterozygous controls (nos 2 and 4). A set of experiments in which *uaY-207* replaced *uaY-9* in the relevant heterokaryons and diploids gave results identical to those obtained using *uaY-9*. In several cases more than one heterokaryon or diploid, originating from parents carrying different forcing markers, were tested. None of these heterokaryons or diploids can utilise hypoxanthine in the presence of 5.5 µM allopurinol.

product in the mycelium or from a genuine nuclear permeability restriction^{4,11-13}. In either case, the phenomenon provides an entirely independent way of demonstrating both that the *uapA* gene is under *uaY* control and that *uap-100* does not alleviate the need for the *uaY* product for *uapA* expression.

Whatever its basis, this nuclear limitation is manifested by a lack of complementation in heterokaryons between *uaY*⁻ mutations and mutations in structural genes under *uaY* control, whilst complementation in the corresponding diploids is clearly observable. As *uap-100* leads to hypersensitivity to inhibition by allopurinol (a substrate of the *uapA* permease^{1,10}) of the utilisation of hypoxanthine (taken up by a different permease²) as nitrogen source¹, it is possible to ask whether *uap-100* can only be expressed when it is in the same nucleus with a functional *uaY* allele. Figure 1 and Table 2 show clearly that *uap-100* is only expressed when in the same nuclei with the *uaY*⁺ allele but not when in nuclei with *uaY-9* and *uaY-207*, even if a *uaY*⁺ allele be present in the other nuclei of equilibrated heterokaryons.

uap-100 is a rather extraordinary mutation; its pleiotropic effects imply that at least a functional overlap exists between a promoter and two initiator sites. It shows a strict dependence on the presence of a functional *uaY* allele but it is expressed in the absence of the ligands of the *uaY* protein. A possible interpretation is that the *uaY* protein is present as two conformers (*P*₁ and *P*₂) in equilibrium, the role of the ligand (co-inducer) being to displace the equilibrium in favour of the form (*P*₂) with the greater affinity for the wild type initiator element. The *uap-100* mutation might decrease the dissociation constant between the receptor site and the conformer favoured in the absence of ligand (*P*₁) or that favoured in the presence of ligand (*P*₂), thus leading to the formation of *uaY* protein-initiator complexes in the absence of inducers. Philip-pides and C.S. (in preparation) have obtained an apparently biphasic curve in equilibrium dialysis experiments with the putative product of the *uaY* gene. This result might reflect the existence of these two conformers of the *uaY* protein.

Further work should reveal whether strict dependence on a functional *uaY* allele is a general characteristic of initiator constitutive mutations for *uapA* gene expression.

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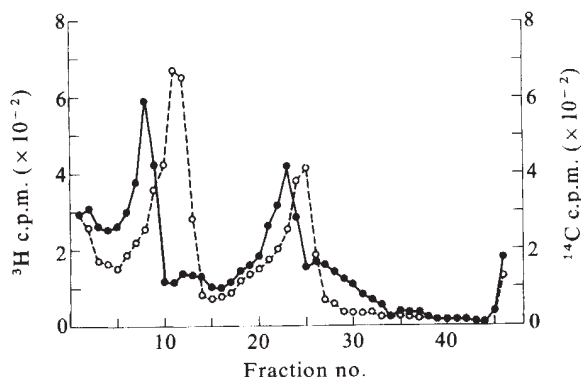
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Transposition of *Pseudomonas* toluene-degrading genes and expression in *Escherichia coli*

PLASMIDS contribute to the metabolic versatility of soil pseudomonads by specifying enzymes responsible for the oxidation of unusual organic substrates^{1,2}. TOL plasmids determine an inducible sequence of enzymes that oxidise toluene, *m*-xylene, and *p*-xylene via alcohol and aldehyde intermediates to benzoate, *m*-toluate, and *p*-toluate, respectively, which are further metabolised by ring fission via the *meta* (or α -ketoacid) pathway—the enzymes of which also plasmid determined³⁻⁵. TOL plasmids found in soil *Pseudomonas* strains⁶ are notably heterogeneous in molecular properties⁷. Analogous observations on the heterogeneity of plasmids carrying the genes for TEM-type β -lactamase⁸ led to the demonstration that this gene was promiscuously transposable between plasmids of Gram-negative bacteria⁹. We have found that *tol* genes can be transposed to other plasmids, including the broad host range plasmid RP4. Such a derivative, RP4-*tol*, allows the expression of a specialised *Pseudomonas* pathway to be studied in *Escherichia coli*. In this foreign host *tol* gene function can be detected but at a much lower level than in *Pseudomonas*.

During studies of the compatibility relationships between the 78×10^6 molecular weight TOL plasmid from *P. putida* (*arvilla*) mt-2⁷ and other *Pseudomonas* plasmids, a derivative of strain AC140 (TOL⁺ *Pseudomonas aeruginosa* PAC) was constructed carrying RP4, a plasmid determining resistance to carbenicillin (Cb), kanamycin (Km), and tetracycline (Tc) that belongs to *Pseudomonas* incompatibility (Inc) group P-1¹⁰ and has a molecular weight of 36×10^6 (ref. 11). Both TOL and RP4 markers were stable and each plasmid could be transferred independently to another *Pseudomonas* recipient, indicating that the plasmids maintained their separate identities. On transfer, however, among transconjugants selected on *m*-toluate as sole carbon source 1 of 26 had lost Tc while retaining Cb and Km. This strain readily transmitted carbenicillin or kanamycin resistance to *E. coli* strain W3110T⁻ (ref. 11). On sucrose gradient centrifugation of purified DNA such transconjugants contained a single plasmid of MW 74×10^6 (Fig. 1).

Fig. 1 Neutral sucrose gradient analysis of plasmid RP4-*tol* DNA. Covalently closed circular tertiary DNA forms of ³H-labelled RP4-*tol* and ¹⁴C-labelled plasmid R1 were isolated using caesium chloride-ethidium bromide density gradient centrifugation¹¹. A mixture of the two plasmid DNAs was analysed by sedimentation through a 4.6 ml 5–20% sucrose gradient for 80 min at 100,000g and 20 °C. Fractions (0.1 ml) were collected and analysed for radioactivity. Sedimentation was from right to left. The molecular weight of RP4-*tol* was calculated as previously described¹¹, using R1 as the sedimentation reference (molecular weight 60×10^6). ●, ³H; ○, ¹⁴C.



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P. aeruginosa strain PU21 recipients¹¹ receiving the plasmid from *E. coli* grew on *m*-toluate, were carbenicillin and kanamycin resistant but tetracycline sensitive, and, like RP4 but unlike TOL, were lysed by donor-specific phages PRR1, PRD1, Pf3, PR3, and PR4, propagated phage G101 poorly, and were tolerant to bacteriocin AR41 (ref. 10). On mating with *P. aeruginosa* recipient carrying IncP-1 plasmid R751 (ref. 10), 0 of 20 carbenicillin-resistant transconjugants and 0 of 20 Tol⁺ transconjugants retained trimethoprim resistance determined by R751, indicating that the *tol* genes had been incorporated into a recombinant, termed RP4-*tol*, that except for Tc retained RP4 functions including IncP-1 specificity.

To determine if the *tol* genes could be translocated from RP4-*tol* to additional plasmids, a transfer-defective derivative was obtained as a survivor following exposure of PU21 (RP4-*tol*) to phage PR4 (ref. 12). IncP-2 plasmid pMG5 that determines sulphonamide (Su) and tobramycin (Tm) resistance among other markers¹⁰ was introduced into this strain, which was then used as a donor selecting for Tol⁺ transfer (Table 1). Most Tol⁺ transconjugants were also carbenicillin resistant as though they had received RP4-*tol* by mobilisation, but 2 of 20 Tol⁺ transconjugants carried only pMG5 markers. These strains uniformly cotransferred the Tol character with Tm whichever marker was used for selection. By Agarose gel electrophoresis a plasmid of MW about 34×10^6 larger than pMG5 was present (J. Hansen and R. Olsen, personal communication) indicating that a pMG5-*tol* recombinant plasmid had been formed by sequential translocation of *tol* genes. Others have also recently described recombination of TOL with plasmids R91 (ref. 13) or RP4 (ref. 14) and of the related XYL plasmid with factor K (ref. 15).

Table 1 Characteristics of Tol⁺ transconjugants

Donor	Frequency of Tol ⁺ transconjugants	Characteristics of transconjugants
PU21 (RP4- <i>tol</i> Tra ⁻)	$<2 \times 10^{-7}$	
PU21 (RP4- <i>tol</i> Tra ⁻) (pMG5)	3×10^{-6}	2/20 Cb ^s Su ^r Tm ^r Tol ⁺

P. aeruginosa PU21 (*ilbB112 leu-1 str^r-1 rif^r*) donor strains carrying a Tra⁻ (transfer-defective) derivative of RP4-*tol* (Cb Km Tol⁺ IncP-1) with or without pMG5 (Km Su Tm Hg IncP-2) were mated with recipient strain PU1 (FP2⁺, prototrophic) for 2 h at 37°C. The frequency of Tol⁺ transconjugants, selected on minimal plates supplied with *m*-xylene vapour as the sole carbon source, was determined relative to the number of donors. Tol⁺ transconjugants were characterised by spotting on antibiotic-containing plates. All transconjugants were Su^r Tm^r; Cb^s transconjugants that could not have received RP4-*tol* by mobilisation were tested as donors. On transfer back to PU21 20 of 20 Tm^r transconjugants were Tol⁺ and 20 of 20 Tol⁺ transconjugants were Tm^r. Cb, Carbenicillin; Km, kanamycin; Su, sulphonamide; Tm, tobramycin; Hg, mercuric chloride.

P. aeruginosa strains carrying RP4-*tol* formed colonies on solid media with *m*-xylene or *m*-toluate in 2–3 days at 30°C. *E. coli* strains carrying RP4-*tol* formed colonies on media with toluene, *m*-xylene, or *p*-xylene as sole carbon sources after 4–5 days. Growth did not occur with alcohol or aldehyde intermediates or with benzoate, *m*- or *p*-toluate although the alcohol and carboxyl substrates were metabolised as indicated by the formation of coloured breakdown products. Several of the toluene and xylene intermediates were toxic for *E. coli*. For example, the alcohol and aldehyde metabolites prevented growth of J53 (RP4-*tol*) with glucose as carbon source. Enzyme assay confirmed that the level of *tol* gene expression in *E. coli* was very low. Oxidation of xylene or xylene metabolites could not be detected with xylene-growth *E. coli* RP4-*tol* using assay conditions in which enzyme activity was readily demonstrable with TOL⁺ *Pseudomonas*¹⁶.

Expression of a *Pseudomonas* catabolic pathway in a foreign host has potential practical application in the development of organisms for industrial use or environmental control. Further studies will be required to determine if the limited expression of *tol* genes in *E. coli* results from a lack of a constituent of the metabolic pathway specified by the *Pseudomonas* chromosome, a preference for the incorporation of *tol* gene products into species-specific membrane components¹⁷, the accumulation of toxic metabolites, or other factors.

Transposability of *tol* genes provides an explanation for the molecular heterogeneity of naturally-occurring TOL plasmids⁷. Excision of a transposable *tol* segment can also account for the formation of Tol⁻ deletion plasmids¹⁸. The minimum contribution of *tol* genes to RP4-*tol* (MW of 74×10^6 minus 36×10^6 , that is, 38×10^6) is far larger than that of known drug resistance transpositions. Formation of *tol*-R plasmid recombinants may involve insertion sequences that flank *tol* genes. In addition to providing a mechanism for translocation, flanking insertion sequences could mediate the reversible alterations in plasmid size observed when certain TOL⁺ strains are grown with *m*-toluate¹⁹, just as insertion sequences flanking r-determinants mediate the dissociation-reassociation of certain R plasmids when their host strains are exposed to antibiotics^{20,21}.

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Identification of a conjugative R plasmid in *Bacteroides ochraceus* capable of transfer to *Escherichia coli*

CONJUGATIVE plasmids specifying multiple antibiotic resistance are commonly found in facultative enteric bacteria. However, R plasmids have not been well characterised in anaerobic organisms, which constitute the major flora of the alimentary tract of mammals. An increasing percentage of anaerobic bacteria isolated from clinical infections are resistant to previously useful antibiotics^{1,2}. Several strains of *Bacteroides* have been reported to contain extrachromosomal DNA, but no function could be attributed to these plasmids³⁻⁶. Recently, Mancini and Behme⁷ demonstrated transfer of antibiotic resistance from *Bacteroides fragilis* to an *Escherichia coli* recipient. However, the individual resistance markers were unstable in the *E. coli* transconjugants, and the presence of closed circular plasmid DNA in either the *B. fragilis* donor or the *E. coli* recipient was not demonstrated. We have previously reported the isolation of two plasmids from a strain of *B. ochraceus*⁵. In the present report the larger plasmid is characterised as a conjugative, multiple antibiotic resistance factor. In addition, it is shown that both plasmids can be transferred to and stably maintained in *E. coli*.

The isolation of *B. ochraceus* strain 2228 and its plasmids have been described previously⁵. This strain produces a penicillinase and is resistant to many antibiotics: penicillin, ampicillin, tetracycline, chloramphenicol, erythromycin, clindamycin, rifampin and the aminoglycosides. Two plasmids were identified in strain 2228, with molecular weights estimated from sucrose gradient sedimentation of 70×10^6 and 25×10^6 (ref. 5). These plasmids have been designated pGD10 and pGD11, respectively. To determine if one or both plasmids was an R factor, we have attempted to transfer antibiotic resistance to an *E. coli* (ref. 8), HB101, was used as the recipient, the selection being for transfer of chloramphenicol resistance. The results of this mating are shown in Table 1. Chloramphenicol-resistant colonies appearing after 24 h of aerobic incubation were shown to be *E. coli* by standard biochemical criteria. Chloramphenicol resistance was transferred to HB101 at a frequency of 4×10^{-4}

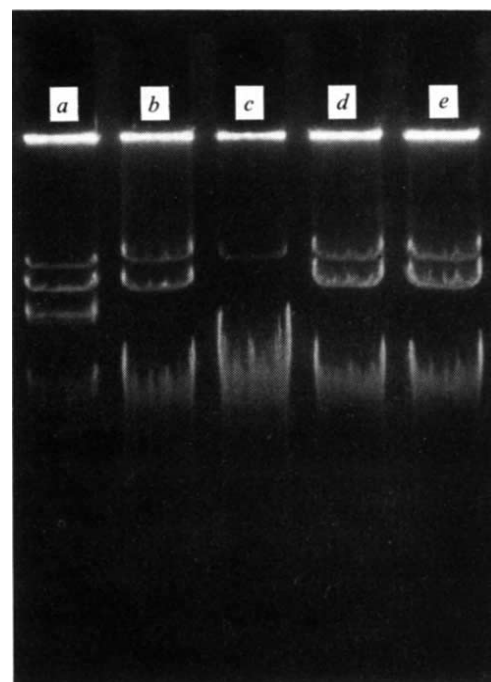


Fig. 1 *B. ochraceus* 2228 and clones of chloramphenicol-resistant *E. coli* HB101 transconjugants were grown separately in 250 ml M9-glucose-casamino acids medium¹¹ to a cell density of approximately 5×10^8 cells per ml. Gentle lysis was induced by successive treatment with lysozyme, EDTA and sodium lauryl sarcosinate, and the plasmid DNA was isolated by caesium chloride-ethidium bromide density gradient centrifugation in a Ti60 rotor as described by Meyer *et al.*¹⁰. The superhelical DNA band was extracted with isopropanol and dialysed against buffer containing 0.1 M Tris-HCl, pH 8.0, 0.05 M NaCl and 0.005 M EDTA. The plasmid DNA was characterised using Agarose gel electrophoresis as described by Meyers *et al.*⁹. a, Marker plasmid DNA species, Rldrd19 (molecular weight 62×10^6 ; ref. 9), RK2 (molecular weight 37.6×10^6 ; ref. 10) and R6K (molecular weight 26×10^6 ; ref. 12). b, *B. ochraceus* 2228. c, d, and e, Individual clones of *E. coli* HB 101 chloramphenicol-resistant transconjugants. Broad bands in the lower portion of the gel represent chromosomal DNA fragments.

Table 1 Frequency of transfer of antibiotic resistance

Donor	<i>E. coli</i> recipient	Resistance markers transferred*	Frequency†
<i>B. ochraceus</i> 2228	HB101	Cm, Tc, Km	4×10^{-4}
Cell-free filtrate of <i>B. ochraceus</i>	HB101	None	0
HB101 (pGD10)	HB101 nal ^r	Cm, Tc, Km	1×10^{-4}

The donor and recipient were grown separately overnight in brain-heart infusion broth (BHI, obtained from BBL). The mating mix contained 1 ml each of the overnight donor and recipient cultures diluted into 10 ml fresh BHI. When *B. ochraceus* was used as the donor, the mating mix was incubated anaerobically in a GasPak jar (BBL) at 37°C. When HB101 (pGD10) was the donor, the mating mix was incubated aerobically at 37°C. After 6 h, the mix was plated on to BHI agar containing 25 µg per ml chloramphenicol and incubated aerobically overnight at 37°C. Colonies were checked for the other resistance markers by replica plating on to BHI agar containing either 25 µg per ml tetracycline or 50 µg per ml kanamycin. The cell-free filtrate of *B. ochraceus* 2228 was obtained by passing the overnight culture of *B. ochraceus* through a Millipore filter (0.45 µm pore size).

* Cm, Tc and Km refer to resistance to chloramphenicol, tetracycline and kanamycin, respectively.

† Frequency of transfer per recipient cell.

per recipient cell. Tetracycline and kanamycin resistance were closely linked to chloramphenicol resistance; all of 300 transconjugant clones initially selected for chloramphenicol resistance were also resistant to tetracycline and kanamycin. The resistance markers were stable in the HB101 transconjugants and showed no spontaneous segregation. The mechanism of transfer was most probably conjugation, as mixing HB101 with a cell-free filtrate of the donor *B. ochraceus* strain resulted in no transfer of resistance (Table 1).

To determine the genetic basis for this resistance transfer, plasmid DNA was extracted from clones of *B. ochraceus* 2228 and chloramphenicol-resistant transconjugants of HB101, and analysed using agarose gel electrophoresis, as shown in Fig. 1. The donor *B. ochraceus* 2228 strain, in well B, contains the two plasmids described previously. In this gel system, the larger plasmid, pGD10, runs slightly slower than the RI marker, as expected for a plasmid of molecular weight 70×10^6 (ref. 9). The smaller plasmid, pGD11, runs coincident with the RK2 marker and on this basis we have revised its molecular weight estimate to 37×10^6 (ref. 10). Three separate clones of HB101 transconjugants are shown in wells C, D and E. One HB101 clone received only pGD10, whereas the other two received both plasmids from *B. ochraceus*. As HB101 containing pGD10 alone acquired resistance to chloramphenicol, tetracycline and kanamycin, these markers could be assigned to pGD10. As

shown in Table 1, HB101(pGD10) was also able to transfer these markers to a naladixic acid-resistant derivative of HB101. Therefore, pGD10 possesses self-transmissible properties. As HB101 is streptomycin resistant, pGD10 was transferred from HB101 to an *E. coli* C600 str^s nal^r strain to determine if streptomycin resistance is specified by this plasmid. The transfer of pGD10 to C600 was followed by selecting for chloramphenicol resistance. All C600 transconjugant clones acquiring pGD10 were also resistant to streptomycin. At present, we cannot assign any genetic markers to the smaller plasmid, pGD11, as *E. coli* strains harbouring this plasmid and pGD10 showed the same antibiotic resistance as the strain containing pGD10 alone. We were unable to detect transfer of resistance to any other clinically useful antibiotic from *B. ochraceus* to *E. coli*. Ampicillin resistance was not transferred to any of the *E. coli* transconjugants tested, suggesting that penicillinase production in *B. ochraceus* strain 2228 is chromosomally determined.

These results identify a conjugative plasmid in *B. ochraceus* strain 2228 which specifies resistance to chloramphenicol, tetracycline, kanamycin and streptomycin. This plasmid can be transferred by conjugation to *E. coli* and is stably maintained in the *E. coli* host. Although our initial studies used a restriction enzyme-deficient *E. coli* recipient, we have recently shown that the presence of the *E. coli* K12 restriction and modification enzyme system does not appreciably reduce the frequency of transfer of pGD10 from *B. ochraceus* into an *E. coli* K12 host. This observation suggests that plasmid transfer may occur *in vivo* between *Bacteroides* and wild-type *E. coli* strains. As *Bacteroides* species are a major constituent of the gastrointestinal tract and mucous surfaces of mammals, they may form a significant reservoir of antibiotic resistance genes.

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Dimethylnitrosamine inhibits enzymatic removal of *O*⁶-methylguanine from DNA

THE induction of tumours by dimethylnitrosamine and *N*-methyl-*N*-nitrosourea is thought to be initiated by the methylation of cellular constituents^{1–3}. Recent results have suggested that a critical product produced by these carcinogens and their ethyl analogues might be *O*⁶-alkylguanine in DNA^{2–5}. Organs relatively resistant to the carcinogenic stimulus provided by these agents seem to have a greater ability to

remove this alkylated purine from their DNA than more sensitive tissues^{5–8}. The activity of the system responsible for the removal of *O*⁶-methylguanine from DNA may, therefore, play an important part in determining whether tumours are produced. The ability to remove *O*⁶-methylguanine from DNA in rat kidney and liver showed a striking change in response to the dose of carcinogen administered. Large single doses of dimethylnitrosamine (20 mg per kg body weight or greater) which produce renal tumours in rats^{1,2} led to almost complete cessation of the excision of this product from kidney DNA⁸. In the rat liver removal of *O*⁶-methylguanine still occurred after high doses of dimethylnitrosamine or *N*-methyl-*N*-nitrosourea but was much less efficient than after smaller doses^{9–11}. All of the studies summarised above were carried out by following the persistence of labelled *O*⁶-alkylguanine in DNA *in vivo* following administration of radioactive *N*-nitroso-compounds. This report describes a comparison of the abilities of extracts derived from liver and kidneys of control rats and rats pretreated with dimethylnitrosamine to catalyse the loss of *O*⁶-methylguanine from acid-precipitable DNA *in vitro*.

Tissue extracts prepared as described in Table 1 legend were incubated with calf thymus DNA which had been alkylated by reaction with *N*-[³H]methyl-*N*-nitrosourea or with rat liver DNA which was alkylated *in vivo* by injection of ³H-dimethylnitrosamine shortly before death. After incubation, the DNA was precipitated with ice-cold 0.25 M perchloric acid, washed and then hydrolysed to release free purine bases which were separated by chromatography on Sephadex G10. Figure 1 shows a typical experiment in which the labelled alkylated bases remaining in the DNA after acid precipitation were measured after incubation in the absence of tissue extract (Fig. 1a), in the presence of extract from normal rat livers (Fig. 1b) and in the presence of a similar amount of extract from livers of rats treated 24 hours previously with 20 mg per kg of dimethylnitrosamine (Fig. 1c). It can be seen that the major peak of radioactivity corresponding to 7-methylguanine was not significantly decreased by the liver extracts. This finding and the observation that the amount of guanine recovered in the hydrolysate of the acid-precipitated DNA was not decreased (see Table 1) show that the extracts do not contain sufficient nonspecific nuclease activity to render a significant amount of the DNA acid soluble under the incubation conditions used. The smaller peak of radioactivity corresponding to *O*⁶-methylguanine was substantially decreased by incubation with the extracts from control rat livers (compare Fig. 1a and b). Therefore, these extracts must contain a specific system for the removal of this product from DNA. However, when the same extracts were prepared from rats treated 24 h previously with a high dose of dimethylnitrosamine the ability to remove *O*⁶-methylguanine was absent (Fig. 1c).

Table 1 shows a quantitative comparison of the activities of extracts from rat liver and kidney to remove *O*⁶-methylguanine from DNA. None of the extracts led to loss of 7-methylguanine or guanine from the acid-precipitable DNA. Liver extracts were more active than kidney extracts in carrying out the removal of *O*⁶-methylguanine but with both liver and kidney extracts the activity was lost if the rats were pretreated with 20 mg per kg dimethylnitrosamine. Mixing experiments indicated that extracts from the nitrosamine treated rats contained an inhibitor of the activity (Table 1). It is possible that part or all of this inhibitor was unlabelled alkylated DNA since these extracts did contain small amounts of DNA such that up to 50 µg of DNA (measured by reaction with diphenylamine¹²) was added. This DNA would be alkylated by the dimethylnitrosamine used in the pretreatment. Since the capacity of the extracts to remove *O*⁶-methylguanine from DNA was limited it is possible that the additional unlabelled DNA was sufficient to dilute the specific activity of the DNA substrate to a point where no activity could be detected. This possibility was tested by purifying the extracts further. As shown in Table 1, experiment 2, the extracts from the nitrosamine-treated rats still lacked activity even after a purification

Table 1 Removal of O^6 -methylguanine from alkylated DNA by liver and kidney extracts

Source of extract	Guanine derivatives present in acid-precipitable DNA		
	Guanine (μmol)	7-Methyl-guanine (p mol)	O^6 -Methyl-guanine (p mol)
Expt 1			
None	2.3	15.8	1.16
Control liver	2.3	15.9	0.40
DMN liver	2.2	16.7	1.12
Control liver + DMN liver	2.4	17.4	1.16
Control kidney	2.4	17.1	0.71
DMN kidney	2.4	17.0	1.09
Expt 2			
None	0.81	11.7	1.38
Control liver	0.72	9.4	0.07
DMN liver	0.73	9.3	1.18
Control liver + DMN liver	0.67	9.1	0.15

Extracts were obtained from livers or kidneys of control rats and rats treated 24 h before death with dimethylnitrosamine (DMN) 20 mg per kg. For experiment 1, the extracts were prepared by homogenisation in 3 volumes of 0.05 M Tris-HCl, pH 7.8, 1 mM dithiothreitol, 0.1 mM EDTA. The homogenate was centrifuged at 10,000 r.p.m. for 5 min at 2°C and the supernatant saved. The pellet was resuspended in 1 volume of the homogenising buffer and sonicated three times for 30 s (Heat-systems model W-225-R, at setting 7). The supernatant was then added and the pellet removed by centrifugation at 15,000 r.p.m. for 30 min at 2°C. The supernatant was then made 80% saturated by addition of solid $(\text{NH}_4)_2\text{SO}_4$ and the resulting pellet collected by centrifugation at 10,000 r.p.m. for 10 min, dissolved in the homogenising buffer and dialysed for 15 h against 50 volumes of the buffer. Each assay contained 15 mg of protein from the extract shown and the other components described in Fig. 1. The substrate ^3H -alkylated DNA was prepared from rat livers removed 30 min after injection of 50 μg per kg ^3H -dimethylnitrosamine ($2.96 \text{ Ci mmol}^{-1}$)¹¹. For experiment 2 the extracts were further purified by chromatography on DEAE-cellulose. After application of the extracts to a column (3 cm $\times$ 20 cm) of DEAE-cellulose equilibrated with the same buffer the column was eluted with a gradient of 0 to 0.3 M NaCl (500 ml) and 10 ml fractions were collected. All of the activity in control extracts eluted in about 8 fractions with a peak at a salt concentration of 0.08 M NaCl and these fractions were collected, concentrated and used as a source of enzyme. The assays contained 5 mg of protein and ^3H -alkylated DNA and other components as described in Fig. 1. The Table shows the amounts of guanine, 7-methylguanine and O^6 -methylguanine remaining in the acid-precipitable DNA after 60 min incubation at 37°C.

step involving chromatography on DEAE-cellulose. No DNA could be detected in the extracts following this step. The inhibitory action towards control extracts observed in mixing experiments was, however, abolished by this purification.

The radioactivity released into the perchloric acid supernatant by incubation of alkylated DNA with tissue extracts was also analysed by chromatography on Sephadex G10. Figure 2 shows analysis of the supernatant fractions from the assays depicted in Fig. 1. There was no sign of any increase in the amount of labelled O^6 -methylguanine present in the supernatant fraction after incubation with control rat liver extracts which led to substantial loss of this product from the DNA. Authentic O^6 -methylguanine was not degraded by the conditions used for preparation of these supernatant fractions. Therefore, this result suggests that the removal may not take place through the action of an *N*-glycosidase as has been observed with extracts from *Escherichia coli*¹³. It is possible that the crude extracts used in these experiments contain further enzyme activities which rapidly degrade all of the released O^6 -methylguanine and this possibility is under investigation. The early eluting peak from the Sephadex column which contains a number of unidentified components

was the only one whose magnitude was increased when the liver extract was present. A small oligonucleotide containing the O^6 -methylguanine would elute in this position but when the supernatant was treated with acid at 70°C in a procedure which would convert such a precursor to the O^6 -methylguanine base, no radioactivity corresponding to O^6 -methylguanine was generated. The label derived from O^6 -methylguanine by the extracts seems to be uncharged and volatile which would be consistent with its being methanol. This could be derived by a direct dealkylation of the DNA substrate or by further metabolism of some other intermediate.

The radioactivity observed in the perchloric acid supernatants which runs between the initial peak and 7-methylguanine was seen in all cases including where incubation was carried out in absence of protein (Fig. 2a-c). This radioactivity and the 7-methylguanine released are probably due to the spontaneous loss of bases such as 7-methyladenine, 3-methyl-

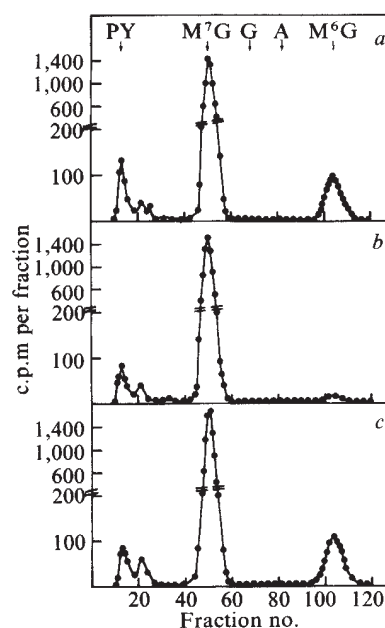


Fig. 1 Separation of labelled purines from acid-precipitable DNA after incubation with liver extracts. An assay mixture (6 ml) containing 33 mM Tris-HCl pH 8, 1 mM dithiothreitol, 3 mM MgCl_2 , [^3H -alkylated]-DNA and 15 mg of extract protein (prepared as described in Table 1, experiment 1) was incubated for 60 min at 37°C. The reaction was halted by the addition of 2 ml of cold 1 M perchloric acid. The resulting precipitate was collected by centrifugation at 0°C, washed twice with 10 ml of 0.25 M perchloric acid at 0°C and then hydrolysed at 70°C in 0.1 M HCl for 30 min to liberate free purines¹⁰. The purines were separated by chromatography on Sephadex G10. Fractions of 5 ml were collected and the radioactivity determined¹⁰. The amounts of guanine and adenine in the hydrolysate were determined by the absorbance of the fractions containing these products and authentic markers of methylated purines were added to establish their elution position¹⁰. The arrows correspond to the peak of the elution of pyrimidine nucleotides and apurinic acid (PY); 7-methylguanine (M^7G); guanine (G); adenine (A); and O^6 -methylguanine (M^6G). a, Separations of purines derived from DNA incubated with no extract; b, extract from normal rat liver; and c, extract derived from livers of rats treated 24 h before death with 20 mg per kg dimethylnitrosamine. The DNA used as substrate was prepared by reaction of 50 μM N -[^3H]methyl-*N*-nitrosourea ($1.055 \text{ Ci mmol}^{-1}$) with calf thymus DNA (5 mg ml^{-1}) in 0.04 M sodium phosphate buffer pH 8.0 for 1 h at 37°C. The DNA was then precipitated by addition of 2 volumes of 2-ethoxyethanol, washed five times with cold ethanol, once with ether and dried in a vacuum for 24 h.

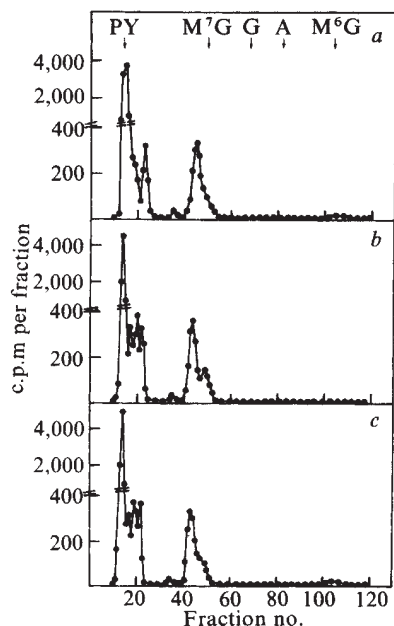


Fig. 2 Chromatographic separation of radioactive products liberated from alkylated DNA by liver extracts and by acid precipitation. After removal of the precipitate formed by addition of perchloric acid to the assays described in Fig. 1 the supernatant was neutralised by addition of 6M KOH. The resulting KClO_4 was removed by centrifugation and the supernatant applied to Sephadex G10 columns and eluted and counted as described in Fig. 1. Radioactivity profiles corresponding to *a*, incubation without protein; *b*, protein from control liver; and *c*, protein from livers of rats treated with 20 mg per kg dimethylnitrosamine 24 h before death.

adenine and 7-methylguanine from DNA on exposure to the acid used for precipitation. 7-Methylguanine is lost from DNA by an acid-catalysed depurination^{4,5}. The amount lost in the experiments described here amounted to less than 2% of that present but the loss of 7-methyladenine and 3-methyladenine in a similar reaction occurs more rapidly^{4,5} and virtually all of these bases were released into the supernatant without the presence of protein in our experiments. For this reason, the possible ability of the extracts to catalyse the loss of these adenine derivatives has not yet been determined.

Our findings confirm the supposition based on the decline in the levels of O^6 -methylguanine in DNA *in vivo*⁵⁻¹⁰ that liver and kidney contain an active enzyme system capable of removing O^6 -methylguanine from DNA and suggest that the reduced efficiency of this process after high doses of alkylating carcinogens may be due to an inhibition of the enzymatic system. The mechanism by which this inhibition is brought about is not due to a general cell necrosis because it occurs more rapidly and to a much greater extent than the cell damage produced by dimethylnitrosamine. This inhibitory action may be important in the induction of tumours by high doses of these nitroso-carcinogens as in the production of renal tumours by single doses of dimethylnitrosamine. The removal of O^6 -methylguanine from kidney DNA is almost completely inhibited within a few hours of such doses⁸. It seems, therefore, that the system which is studied here may be the only process for O^6 -methylguanine removal from DNA in the kidney. In the liver the activity of the present enzyme system was also completely inhibited by pretreatment with similar doses of dimethylnitrosamine but O^6 -methylguanine removal from DNA *in vivo* continues although at a much less rapid rate than after low doses of the carcinogen⁸⁻¹¹. Therefore, the liver cells may also contain an additional process for the removal of

O^6 -methylguanine from DNA which is not subject to this inhibition.

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Electron microscopic location of protein thiol residues

ATTEMPTS to use conventional transmission electron microscopy to visualise single heavy atoms attached to protein thiol residues are usually unsuccessful, because the signal due to the heavy atom is generally too weak to be detected against background noise and the signal due to the protein and negative stain (if used). This is unfortunate because such a technique would prove invaluable as a means of determining molecular orientations in multiprotein assemblies and relative positions of thiol residues within individual molecules. As one approach to this problem we have devised and report here a method by which the signal at the thiol residues can be increased by introducing several heavy atoms at each site. This considerably aids ultrastructural localisation by electron microscopy, particularly in the presence of heavy metal negative stains such as uranyl acetate. We illustrate the technique by visualising the thiol residues in uranyl acetate-negatively stained magnesium paracrystals of rabbit skeletal muscle tropomyosin. Although the conditions we report gave satisfactory results with tropomyosin, they should only be considered as a guide when labelling other proteins, as the optimum reaction conditions may be different in some cases.

The method exploits the well known property of pyrrole rings to bind up to four mercury atoms¹ and entails a two-stage reaction sequence (Fig. 1). A pyrrole ring is first introduced at each protein thiol residue by reaction with *N*-pyrroloisomaleimide (NPIM), to give the pyrrolosuccinimido adduct (II) by addition and rearrangement². Subsequent treatment with HgCl_2 results in four mercury atoms being introduced into each pyrrole ring. Although reaction does not normally take place at disulphide bonds, these could be labelled as effectively as thiols if they were first reduced. The magnesium paracrystals of rabbit skeletal muscle tropomyosin, a rod-shaped α -helical protein³, are a convenient test object because they give a

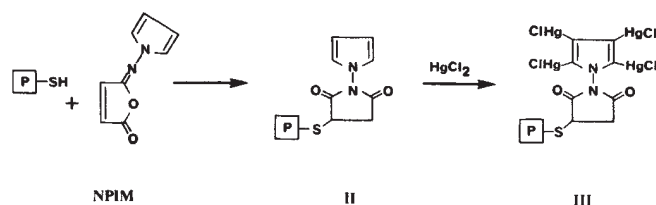


Fig. 1 Reaction scheme used to introduce multiple mercury atoms at each thiol residue. The protein, P , is first modified with *N*-pyrroloisomaleimide (NPIM) to produce the protein-pyrrolosuccinimido adduct (II), which is subsequently treated with HgCl_2 , whereby four mercury atoms are introduced into each pyrrole ring. The structures of the adduct (II) and its mercurated product (III) are consistent with the stoichiometries observed and are based on the results obtained on model compounds^{1,2}, as the small quantity of the product and the large amount of protein present precluded a definitive chemical analysis.

simple pattern of bands when negatively stained³, and also because the location in the paracrystals of the single thiol of α -tropomyosin (which comprises about 75% of rabbit skeletal tropomyosin⁴; the minor (β) component has an additional thiol residue at an unknown location) has already been established⁵.

In common with the closely related protein thiol modification reagent *N*-ethylmaleimide (NEM), NPIM reacts with both thiol and amino groups², but it is possible to restrict the reaction mainly to the thiol residues by using appropriate reaction conditions. Because the selectivity of this reaction is usually enhanced in acidic conditions⁶ (due to protonation of the amino group), we carried out the modification reaction in 50 mM Na acetate buffer, pH 5.5, at room temperature, and, because the thiols of the native protein were inaccessible, we also added urea to denature the tropomyosin. (When the urea is removed, tropomyosin regains its original conformation⁵, but this is not the case for all proteins. Consequently, we would advise the addition of urea only when adequate reaction cannot be obtained in the native state and when it is known that the protein can be successfully renatured.)

We assessed the selectivity of the modification procedure by examining the kinetics of reaction with tropomyosin in which the thiol residues were alternatively free (reduced) or blocked (by disulphide formation with dithio-bis-(nitrobenzoic acid), DTNB), both by measuring the rate at which the NPIM absorbance at 345 nm decreased and the rate at which pyrrole groups were incorporated into the protein. Both techniques indicated that the protein thiol reacted about 400 times faster than the amino groups, but this is, of course, partially counterbalanced by the higher concentration of the latter (39 amino groups per thiol in tropomyosin⁴). We observed a similar selectivity when reacting NPIM with amino acids and do not anticipate that it would vary greatly between proteins, provided that their thiol residues were sufficiently exposed to react freely. In some instances, however, steric factors could influence the reaction and so we caution that the selectivity should always be checked (for example, by determining the degree of incorporation of NPIM when the thiols are, respectively, blocked and free). The rate of incorporation of NPIM into both the reduced and oxidised protein is shown in Fig. 2, where it is clear that, although reaction does occur in addition to that at the thiol residue, its extent is small by the time most of the thiol has been modified. On the basis of these kinetic results, it was decided to use a twofold excess of NPIM over thiol and a reaction time of 12 min as standard conditions, whereby it was anticipated that almost all the thiol residues would have reacted, and the labelling of amino groups should not exceed 0.1 residues per labelled thiol. In these conditions, α -tropomyosin and unfractionated (that is, the natural mixture of α and β) tropomyosin took up, respectively, 1.10 ± 0.08 and 1.07 ± 0.08 mol of NPIM per mol of thiol (mean $\pm$ s.d.).

Mercuration of specimens for electron microscopy was most conveniently carried out on samples attached to carbon films (see legend to Fig. 3) to prevent aggregation. Because of the small quantities used, it was not possible to estimate the amount of mercury bound to these specimens, but studies on larger samples mercurated in solution indicated that four mercury atoms would be bound to each pyrrole ring. As a control we used α -tropomyosin modified with NEM (which gives a protein-succinimido adduct in which the pyrrole ring in II is replaced by an ethyl group). Samples containing 1–2 mg of magnesium paracrystals were treated with 0.2 M HgCl_2 overnight and then dialysed against 2×100 volumes of 50 mM MgCl_2 , 50 mM Tris-HCl, pH 8, at 4°C followed by 100 volumes of 1.5 M KCl, 25 mM Tris-HCl, pH 8, at 4°C. In these conditions 3.1 ± 0.2 atoms of Hg per cysteine (assayed by atomic absorption spectrometry) remained bound to NEM-modified tropomyosin (this is not surprising as tropomyosin has a strong tendency to bind divalent cations). By contrast, NPIM-modified tropomyosin retained 7.8 ± 0.4 Hg atoms per cysteine, an increase of 4.7 ± 0.3 per cysteine or 4.3 ± 0.5 per pyrrole ring.

The position of the thiol residues was clear in micrographs of both unstained and uranyl acetate-negatively stained paracrystals (Fig. 3). As a control we used paracrystals of α -tropomyosin modified with NEM, and when mercurated then negatively stained with uranyl acetate, they showed the characteristic pattern of bands normally observed in tropomyosin paracrystals (Fig. 3a)^{7,9}. The molecules in these paracrystals lie antiparallel⁷ with their C-termini overlapping by about two-thirds of the molecular length^{5,8,9}. These overlaps correlate with the alternating series of light and dark zones seen in negatively stained paracrystals: the 27-nm light zones cor-

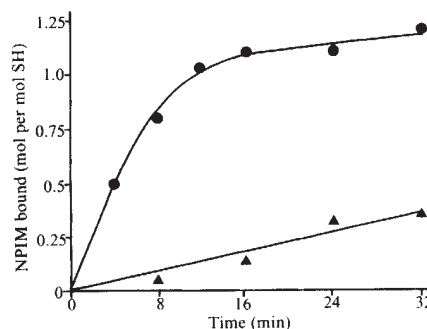


Fig. 2 Rate of formation of protein-pyrrolosuccinimido adduct (see Fig. 1). Rapid reaction occurs (●) when tropomyosin is reduced (by dialysis against 100 vol 15 mM 2-mercaptoethanol, 1 mM EDTA, 50 mM Tris-HCl, pH 8 at 4°C, followed by 4×100 vol of 5 mM HCl to remove excess 2-mercaptoethanol¹⁰) but not when the tropomyosin thiols have been blocked by disulphide formation (▲) by reaction with DTNB as described in ref. 11. The reaction of protein thiol in the reduced tropomyosin has neared completion after about 12 min. A 5–10-mg ml^{-1} solution of either reduced or oxidised tropomyosin (prepared as in ref. 9) was made up in 6 M urea, 50 mM Na acetate, pH 5.5, at room temperature, and the modification reaction initiated by adding a twofold excess over thiol of NPIM (prepared as in ref. 2) as a 2% solution in dimethylsulphoxide (DMSO). Samples were taken at various time intervals and quenched by adding an approximately 20-fold excess of cysteine over NPIM. NPIM not bound to the protein was removed by dialysis against 25 mM Tris-HCl, pH 8, at 4°C. The quantity of NPIM bound was then assayed by adding 0.2 ml of the protein solution to 3 ml of 2.5% 4-*N,N*-dimethylaminobenzaldehyde in 1.5 M H_2SO_4 , heating the resultant solution at 70°C for 7 min, reading the optical density at 550 nm and comparing it with a series of NPIM standards of known concentration. Protein concentration was measured by the method of Lowry *et al.*¹² using bovine serum albumin as standard.

respond to the overlap of the C-termini (CC zone), and the 13-nm dark zones correspond to the overlap of the N-termini (NN zone)^{8,9}. When the protein was modified with NPIM and mercurated, a prominent new dark band appeared in the middle of the CC zone. This band is due to the mercury atoms attached to the pyrrole ring of the thiol-succinimido adduct and is in the position known to be occupied by the thiol residues in these paracrystals⁵. In the absence of uranyl acetate, the control (NEM/Hg) paracrystals are essentially featureless, but those modified with NPIM/Hg again display a very clear pattern of bands at 40-nm intervals. The random background pattern of dots probably represents NPIM bound to amino

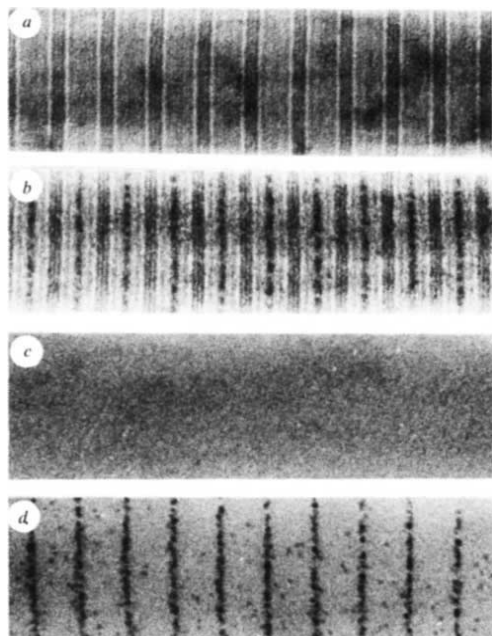


Fig. 3 Electron micrographs of magnesium paracrystals of α -tropomyosin (all at magnification $150,000\times$). *a*, Modified with NEM (using the reaction conditions as for NPIM), mercurated, then negatively stained with uranyl acetate. The pattern can be divided into alternating 27-nm, lightly staining zones and 13-nm, darkly staining zones. *b*, Modified with NPIM, mercurated, then negatively stained. Note the appearance of the densely staining band (which indicates the position of the thiols) in the middle of the lighter zones. *c*, Modified with NEM, mercurated, unstained. The pattern is featureless. *d*, Modified with NPIM, mercurated, unstained. Note the clear single line every 40 nm which indicates the position of the thiol. The protein was modified by treating a $2\text{--}5\text{ mg ml}^{-1}$ solution of α -tropomyosin (prepared as in ref. 9) in 6 M urea, 50 mM Na acetate, pH 5.5, at room temperature with a twofold excess (based on thiol) of either NEM or NPIM as a 2% solution in DMSO. After 12 min the reaction was terminated by the addition of an approximately 20-fold excess of cysteine and the protein renatured by dialysis against 2×100 vol of 25 mM Tris-HCl, pH 8, at 4°C . Paracrystals were formed by dialysis against approximately 100 volumes of 50 mM MgCl_2 , 50 mM Tris-HCl, pH 8 at 4°C . Paracrystals were formed by dialysis prepared by floating thin (about 10 nm) evaporated carbon films directly from mica onto the surface of the paracrystal suspension and then washing with other solutions as required: mercuration was carried out by floating the film overnight on 0.2 M HgCl_2 followed by 50 mM MgCl_2 , 50 mM Tris-HCl, pH 8, at 4°C for at least 30 min to remove mercury nonspecifically bound; negative staining was carried out by floating the film on 2% aqueous uranyl acetate for several minutes followed by 10 s on distilled water to remove excess stain. Unstained specimens were more stable under the electron beam when floated on 1% aqueous glutaraldehyde followed by distilled water (each for about 10 s). After carrying out the required treatments, the film was picked up on 400-mesh copper grids and examined in standard conditions in a Philips EM301 electron microscope.

groups. Because this pattern is both weak and random it does not interfere with the interpretation. No significant interference was detected from Hg bound to sites other than the pyrrole rings (Fig. 3c), either because its signal was too weak or because it was distributed over several sites.

In both unstained and negatively stained paracrystals, the NPIM/Hg band was sufficiently stable under the electron beam to allow focusing and normal photography and was sufficiently intense to be easily visible by eye on the main viewing screen. This is in marked contrast to the results obtained when only a single Hg was introduced at each thiol⁵. In this instance, the Hg signal was too weak to be detected in the presence of uranyl acetate and could only be detected in unstained material in special conditions and after using sophisticated computer image processing techniques⁵.

These results demonstrate that NPIM can be used to label selectively protein thiol residues sufficiently densely to allow their visualisation by conventional transmission electron microscopy. Of particular importance is the observation that the stain is sufficiently dense and stable under the electron beam to allow its detection in normal operating conditions, without having to use sophisticated imaging (for example, low dose, dark field or scanning transmission) or computer or optical processing techniques which are generally not available in a routine electron microscope facility. We anticipate that this technique will prove particularly effective in determining the positions of subunits in multiunit systems (for example, the present result confirms the absolute orientation of the molecules in the paracrystals and so locates the troponin binding site about one-third along the molecule from the C-terminus^{5,11}) and determining the relative position of thiol residues within protein molecules.

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matters arising

Oblique spreading and fracture zones

REARRANGEMENT of the data on fracture zones in the North Atlantic leads to a different answer to the question raised by Atwater and Macdonald¹: "Are spreading centres perpendicular to their transform faults?". They conclude that in slow spreading areas, like the North Atlantic, the spreading is oblique. We, however, suggest that two classes of transform faults exist—orthogonal (or near-orthogonal) and oblique. The former class has large offsets, the latter class is characterised by offsets of the order of 20 km. This relationship is shown in Table 1.

Given these two classes of transform faults, we have to decide which one represents the seafloor spreading direction. In this context we note that Searle and Laughton² describe the Kurchatov FZ as a zone of oblique spreading and

Table 1 Characteristics of orthogonal (class I) and oblique (class II) transform faults

Location	Name	Angle of obliqueness	Offset
Class I			
10°50'N	Vema FZ	2°(9°)	290 km
12°10'N		~0°	45 km
12°40'N		~0°	80 km
15°20'N	Fifteen twenty FZ	3°(7°)	165 km
23°45'N	Kane FZ	0° (south)	130 km
30°00'N	Atlantis FZ	6°	40 km
35°15'N	Oceanographer FZ	0°	165 km
Class II			
13°45'N	Fracture Zone C	30°–40°	16 km
16°40'N		30°–40°	18 km
17°40'N		30°–40°	~20 km
36°15'N		17°	18 km
36°37'N		17°	18 km
36°56'N	Fracture Zone B	17°	18 km
40°40'N	Fracture Zone A	17°	18 km
	Kurchatov FZ	35°	18 km

The data between 12° and 18°N result from a recent survey (B. J. C. *et al.*, in preparation). Where our figures differ from Atwater and Macdonald's listing, their values have been added in parentheses.

shearing. This type of transform would occur in fracture zones with short offsets in preference to 'true' transform faults. The explanation is that the asthenospheric conduit is not interrupted over such short distances (~20 km).

This description applies also to the 13°45'N FZ which, like the Kurchatov FZ, has no regular fossil extension but shows as an irregular zigzagging zone in the topography of the ridge flank. For the area between 12° and 18°N we therefore conclude that the spreading direction is defined by the orthogonal system of larger offset transform faults and median valley segments (Fig. 1a). We propose a similar interpretation for the FAMOUS area where the Oceanographer FZ would represent the spreading direction and Fracture Zones A, B and C are of the oblique spreading type (Fig. 1b). This explains the absence of signs of adjustment to a new spreading direction³ near the Oceanographer FZ and is also in accordance with the circumstance that Fracture Zones A, B and C cannot be followed in the flank topography.

We left out the Charlie-Gibbs FZ from Table 1. To the north (but not directly adjoining this fracture zone) indeed 'true' oblique spreading occurs, that is, no kinematic solution can be formulated without assuming an anomalous behaviour of Reykjanes Ridge. However, for the central North Atlantic our answer to Atwater and Macdonald's question is

that we have no evidence for oblique spreading in recent times other than in very narrow zones, the class of small offset fracture zones.

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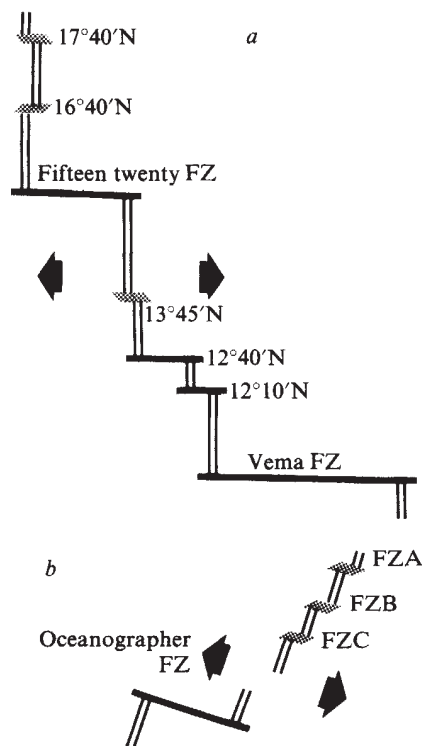
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ATWATER AND MACDONALD have demonstrated that slow-spreading mid-ocean ridges may trend obliquely to the spreading normal, not perpendicular to it¹. They suggest that the angle between ridge-trend and spreading normal may be as great as 35°–38° in two places on the Mid-Atlantic Ridge: Kurchatov FZ (41°N) and Charlie-Gibbs FZ (53°N). I believe that these two instances may be misleading as in each case it is only a short segment of ridge which displays a highly oblique trend, whereas adjacent ridge segments are nearly orthogonal to the spreading direction.

The Kurchatov FZ case was reported by myself and Laughton². We believe this to be a case of oblique spreading occurring in a fracture zone instead of transform faulting: the oblique-spreading

Fig. 1 Geometry of the Mid-Atlantic Ridge between 10°50' and 18°N (a) and between 35° and 37°N (FAMOUS area) (b). Arrows indicate proposed spreading directions.



segment joins directly to normal-spreading ridge segments to its north and south (Fig. 1a), so this case does not correspond to any of those considered by Atwater and Macdonald (their Fig. 1).

In June and July 1977 *RRS Discovery* made a survey of part of Charlie-Gibbs FZ using the long-range sonar GLORIA. I have recontoured the available bathymetric data using the sonar results for control, and also incorporated some unpublished results from the Deep-Tow survey (made available by P. Lonsdale). The resultant charts show that the spreading axis south of the fracture zone is completely orthogonal ($92^\circ \pm 2^\circ$) to the fracture zone trend. On the north side of the fracture zone, the spreading axis is indeed highly oblique to the spreading direction, as reported by Atwater and Macdonald, but probably only over a distance of some 30–40 km from the fracture zone (Fig. 1b). From about 53° to 56° N the spreading axis is at least roughly orthogonal to the spreading direction.³

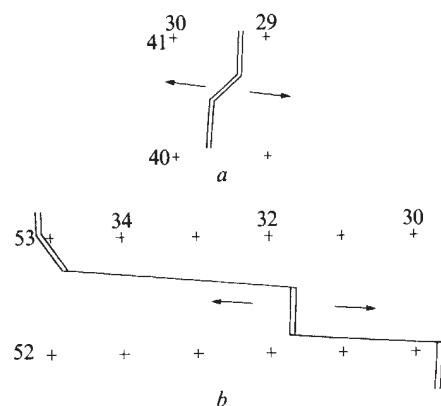


Fig. 1 Spreading axes (double lines), transform faults (single lines), and spreading directions (arrows) at a, Kurchatov FZ and b, Charlie-Gibbs FZ.

Thus there are apparently two types of oblique spreading: a general obliquity, up to about 18° , on slow-spreading ridges, and a stronger obliquity of about 40° near some fracture zones. In the latter case, the obliquity corresponds to the trend of tension cracks produced in initial shearing of virgin material⁴, suggesting that oblique spreading near fracture zones results from the response of the lithosphere to local shearing².

Sea-floor lineations (probably fault-scarps) trending about 40° from the spreading normal have been observed in all the fracture zones studied with GLORIA⁵, as well as Siqueiros FZ on the fast-spreading East Pacific Rise⁶ and in Afar, North-east Africa⁷. However, the extent to which oblique spreading develops obviously varies, as evidenced by the fact that it extends 30–40 km on the north side of Charlie-Gibbs fracture zone but is negligible on the south side. This development may therefore depend

critically on factors such as local crustal thickness or lithospheric strength (thicker or stronger lithosphere would distribute mantle-derived shear stresses over a wider area), or possibly relative motion between the plates and the underlying asthenosphere in directions parallel to the ridge axis (the 'leading edge' of a fracture zone in such an environment would experience only divergent motion, whereas the 'trailing edge' would be subjected to shear persisting in the asthenosphere after the fracture zone had passed over it).

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ATWATER REPLIES—In our paper we discussed the relationship between transform faults and nearby spreading centres. In fact, it is more interesting to address two related but more fundamental questions. First, are all transform faults parallel to the relative motion of their adjacent plates? And second, are spreading centres perpendicular to that motion? Because of the ambiguities surrounding the Oceanographer Fracture Zone, the relative motions between the African and North American plates are uncertain, so that we were unable to consider these questions in the FAMOUS area.

Although Collette and Sloomweg only consider spreading centre–transform relationships, their comments suggest that in fact the centres may be nearly perpendicular to the plate motion direction, while the trend of transform faults may or may not be parallel to that direction, depending on the length of offset. Since their discussion is based on unpublished data, we cannot comment at present, except to say that this is a very important and interesting observation, if true. The region studied by Collette and Sloomweg is especially appropriate for consideration of this question because the Vema FZ is a very long, clean cut fracture zone, probably a dependable indicator of plate motion direction. We look forward to the publication of their data. We shall be especially interested in examining their track layout over the features of interest. In our paper we had to reject several surveys for lack of sufficient resolution of the small angular differences we were measuring.

Both respondents commented on the obviously anomalous behaviour of the Kurchatov FZ. Although we included it for completeness, we agree that its direction seems to be spurious. To include it with the FAMOUS fracture zones A and B, however, may be a mistake, since the trends of the latter are consistent from one to the next and are closer to the spreading direction. Indeed, if the more southeasterly trend of the Oceanographer FZ is taken, as Collette and Sloomweg suggest, the obliquity at the FAMOUS fracture zones is reduced to 5 – 10° .

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Benthic nutrient regeneration and high rate of primary production in continental shelf waters

ROWE *et al.*¹ have recently contended that nutrient regeneration in sediments is the major factor responsible for the relatively high rate of primary production observed in continental shelf waters. They state: "If there were no contribution (of NH_4^+) from the bottom sediments the system would lose this feedback and the production of the plant biomass, dependent only on water column regeneration, would be reduced to rates similar to those found beyond the continental shelf." The contention of Rowe *et al.* is based primarily on two tenets: first, that there is a gradient of decreasing NH_4^+ concentration between the benthos and overlying water; second, that NH_4^+ release can be estimated from measurements of respiration in bottom sediments. We feel that on the continental shelf the sediments play a minor part in cycling nitrogenous nutrients for phytoplankton.

It is clear from several studies (refs 2–4 and L. Conway, personal communication), that an above-bottom NH_4^+ gradient is not a ubiquitous feature of the northeastern United States shelf. As there is great lateral variability in NH_4^+ concentration on the shelf, together with a mean water current velocity of about 5 to 6 cm s^{-1} (ref. 5), the horizontal advection of sediment-generated NH_4^+ must be much greater than the vertical; and therefore this latter term must be lower than the authors' estimate.

The NH_4^+ flux rates could not be as high as claimed, as there is not enough energy available to support the required benthic metabolism. Using three of Corwin's⁴ NH_4^+ profiles from over a 'typical sand bottom' on the shelf south of Long Island, Rowe *et al.* calculated

a flux of $144 \mu\text{g-atom NH}_4^+ \text{ m}^{-2} \text{ h}^{-1}$. This rate would require the oxidation of $500 \text{ mg C m}^{-2} \text{ d}^{-1}$. As phytoplankton production averages only $400 \text{ mg C m}^{-2} \text{ d}^{-1}$ in this area⁶ and the majority of this energy is utilised by zooplankton and nekton in the water column, it is impossible that the actual rates of NH_4^+ regeneration could have been this high unless there was a large supply of allochthonous organic material to the sediments. Being sandy sediments, the input of allochthonous material must have been low.

It seems that the authors may have overestimated the value and constancy of the NH_4^+ release ratio. In calculating NH_4^+ release from benthic respiration data, they assume that for each millilitre of O_2 consumed by the sediment, 0.041 mg NH_4^+ are produced and that 81% of this is released to overlying waters. In Narragansett Bay the release of NH_4^+ averaged only 60% of that predicted from atomic ratios⁷, and in a Scottish sea loch there was a 15-fold variation in the observed O_2 consumption: NH_4^+ evolution ratios⁸.

In contrast to the continental shelf, it seems that benthic nutrient regeneration is important in some nearshore areas. In Narragansett Bay, high (that is, $50\text{--}300 \mu\text{g-atom NH}_4^+ \text{ m}^{-2} \text{ h}^{-1}$) rates have been measured⁷⁻⁹, but in some other shallow areas such as the Chesapeake Bay¹⁰ and off southern California¹¹ the benthos supplies less than 5% of the approximated phytoplankton nitrogen demand.

The deep water at the edge of the continental shelf contains about $15\text{--}25 \mu\text{g-atom NO}_3^- \text{ nitrogen l}^{-1}$, and Riley¹² has suggested that the movement of this water on to the shelf is the most likely source of the nitrogen needed for elevated production in this region relative to off-shore areas. Recent studies¹³⁻¹⁷ support this hypothesis.

In some shallow coastal waters, benthic nutrient regeneration may be a major source of nitrogen; however, the available evidence does not uphold the contention that it supports the relatively high rate of primary production present on the continental shelf.

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ROWE REPLIES—Several years ago we initiated a study of the relationships between benthic biological processes and water column productivity in sublittoral marine environments. Simple numerical models of the conversions of organic matter to inorganic products indicated that high rates of metabolism must be putting large quantities of ammonia and nitrate back into the water column. This seems intuitively obvious because in shallow water most of the biomass is on the bottom, rather than in the water column. Aware that nutrient sources and cycling were of importance to primary productivity, we looked for evidence for our hypothesis that nutrient regeneration in sediments was important. We easily found such evidence, first in the data report of Corwin¹, on which our first paper was based². Later we embellished this hypothesis by contouring the vertical and horizontal distribution of ammonia between New York and Cape Hatteras³. Since the presentation of our model, we have not remained idle. To quantify the importance of sediments as a site of regeneration, we began to collect data on the concentration of ammonia and nitrate in sediment pore waters and to measure their fluxes from the bottom by incubating small areas under bell jars.

Off north-western Africa, in an upwelling area, our measurements suggested that about one-third of the nutrients supplied to the phytoplankton came from bottom regeneration, with the other two-thirds supplied by regeneration in the water column and advection (upwelling)⁴. Our studies off western South America have demonstrated high concentrations of both ammonia and nitrate in interstitial waters (ref. 5 and G. T. R. and N. Staresinic, unpublished), and it is again apparent that bottom regeneration is important. Also we have had the opportunity of studying the New York Bight, the area from which our original data were obtained, on two more recent occasions⁶. (Dr John Walsh heads a group at the Brookhaven National

Laboratory that is conducting continuing investigations on the secondary productivity of the continental shelf off Long Island and New Jersey. We joined them on KNORR cruise 68 in August, 1977.)

In a silty clay of the Christiaensen Basin we measured a regeneration rate of $24 \text{ mg N m}^{-2} \text{ d}^{-1}$ (ref. 6), whereas in the coarse sand along New Jersey and south of Long Island we measured rates that averaged $105 \mu\text{g-atom m}^{-2} \text{ h}^{-1}$, or $35 \text{ mg N m}^{-2} \text{ h}^{-1}$ (range 29-158 $\mu\text{g-atom m}^{-2} \text{ h}^{-1}$, $n=6$). The work in New York Bight with which we are associated⁸ suggests that two bands of chlorophyll are evident on the continental shelf: one nearshore and one along the shelf slope break. We attribute the nearshore one to association with the bottom and bottom regeneration, but the offshore band may result from the 'upwelling' proposed by Riley⁷, referred to by Carpenter and McCarthy.

Carpenter and McCarthy make no *ab initio* suggestions of why productivity is higher on the continental shelf, unfortunately, and they did a poor job of trying to shed doubt on our hypothesis, especially in light of our commensurate studies. Bottom water ammonia gradients are present over the inner half of the shelf when the water is stratified^{2,3,6}.

Our rates of remineralisation (cited by Carpenter and McCarthy) are greater than the yearly average, as our estimates^{2,3} do not include the winter when metabolic rates are extremely low because of low temperatures. How much of the primary productivity is used by pelagic organisms before it gets to the bottom is not quantified for the New York Bight, but indirect evidence that little is used in the water column is supported by the work of our collaborators continuing there.

Given experiences listed above, we are confident² that nutrient regeneration occurs extensively in continental shelf sediments. We agree, however that its importance to productivity remains hypothetical. Regeneration on the bottom as opposed to the water column may increase productivity because ammonia is a favoured substrate for phytoplankton⁸, and it is the dominant form of inorganic nitrogen (besides N_2) in sediment pore waters.

Sediments may also act as a reservoir that is much easier to tap than deep water offshore. We continue to believe that somehow the structure and functioning of the shelf ecosystem may be different from an ocean far from its bottom, and that a manifestation of this difference may be the shelf's somewhat higher productivity.

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NH₄Cl and other weak bases in the activation of sea urchin eggs

TWO recent reports^{1,2} have demonstrated the importance of intracellular alkalinisation in initiating development of sea urchin eggs. However, in accounting for the ability of certain weak bases to initiate development by raising intracellular pH (pH_i), the authors used *ad hoc* assumptions, where a straightforward physiological explanation would have sufficed.

Loeb³ first showed that ammonia can trigger development of unfertilised sea urchin eggs. In 1976, Johnson *et al.*¹ observed that pH_i rises rapidly in the first minutes after fertilisation, and showed that this rise is associated with both an efflux of acid (fall in pH_o) and the influx of an equivalent amount of Na⁺. Egg development and the changes in pH_i and pH_o could be prevented both by amiloride and by incubation in Na⁺-free media. The authors concluded that fertilisation activates an amiloride-sensitive Na⁺:H⁺ exchange mechanism which raises pH_i , thereby causing the metabolic derepression that initiates development. More recently Lopo and Vacquier² showed that the rapid initial pH_i rise is followed by a slower fall which is largely prevented by 2,4-dinitrophenol (DNP).

In addition, Johnson *et al.*¹ found that certain weak bases (ammonia, nicotine, procaine), when added to fertilised eggs in Na⁺-free sea water, even in the presence of amiloride, caused the characteristic changes in pH_i and pH_o and initiated development. The authors made the *ad hoc* assumption that these pH changes were caused by the amines either inducing only one component (that is, H⁺ efflux) of the Na⁺:H⁺ exchange, or substituting directly for Na⁺. Lopo and Vacquier² showed that procaine, known to reversibly stimulate DNA synthesis in unfertilised eggs⁴, also causes a reversible increase in pH_i . They resorted to the explanation that procaine raises pH_i by increasing membrane permeability to H⁺. We propose to replace these *ad hoc* assumptions by a simpler explanation, namely, that the weak base enters the cell as the uncharged molecule (B), which then combines with protons to yield the charged form (BH⁺). The authors discarded this mechanism, apparently

because they considered it incompatible with the observed fall in pH_o . However, such a fall is precisely what would be expected. The equilibrium $BH^+ \rightleftharpoons B + H^+$ in the medium shifts to the right as B leaves the medium for the cells, thereby lowering pH_o . The entering B simultaneously raises pH_i , as has been demonstrated in several cells with pH-sensitive microelectrodes⁵⁻⁷. These changes in pH_o and pH_i can thus occur in the absence of the transmembrane movement of H⁺ *per se*, and clearly should be neither Na⁺-dependent nor amiloride-sensitive. When the weak base is removed from the bathing medium, B leaves the cell, the above reactions reverse, and both pH_o and pH_i return towards their initial values. Indeed, Lopo and Vacquier observed such a fall in pH_i . It follows that this fall in pH_i is DNP-insensitive as these authors found. We propose that weak bases circumvent the normal mode of raising pH_i in sea urchin eggs by directly penetrating the cell membrane and consuming protons.

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EPEL ET AL. REPLY—Fertilisation initiates a defined sequence or programme of events and one of these, the massive extrusion of protons referred to as the fertilisation acid, was described almost 50 years ago¹. Ammonia (and other amines) acts like a parthenogenetic agent in turning on certain 'late' events of this sequence, such as K⁺ transport², protein³ and DNA synthesis^{4,5}, and in fact its mode of action was postulated as a pH effect^{2,3}. The subsequent finding that ammonia mimicked fertilisation in causing a rapid acidification of the seawater when added to eggs⁶ led us to assume that ammonia and the various amine compounds were inducing yet another step of the fertilisation sequence, that is, the fertilisation acid. This discovery, of course, led to the subsequent descriptions of the Na⁺-H⁺ exchange and the resultant alkalinisation of the cytoplasm which seems essential for biosynthesis⁷. The explanation for the external acidification that is proposed by Boron *et al.* above, and which has since been confirmed experimentally by Winkler and Grainger⁸, provides a simpler explanation and we concur with them.

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Nitrogen-fixing root nodules in Ulmaceae

A SURVEY of symbioses of *Rhizobium* spp. with members of the Ulmaceae and Urticaceae in Java and Bali (Indonesia)¹ has revealed nitrogen-fixing nodules only in *Parasponia parviflora* Miq. (Ulmaceae). None were found in specimens of *Trema*, which are morphologically closely related to *Parasponia* spp. The survey has shown that reports that nodulated specimens of the Ulmaceae from Papua New Guinea belonged to *Trema aspera*² or *T. cannabina* Lour. variety scabra (Bl.) de Wit³ were incorrect. The specimens have now been shown to belong to *P. rugosa* Bl. We believe that *Trema* and *Parasponia* spp. have been confused frequently in the past. Symbioses of *Rhizobium* spp. with members of the Ulmaceae probably occur only in the Asiatic genus *Parasponia*.

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reviews

Processes of scientific intelligence

Edward Bullard

Most Secret War: British Scientific Intelligence, 1939-1945. By R. V. Jones. Pp. 542. (Hamish Hamilton: London, 1978.) £6.95. (Published as *The Wizard War* by Coward, McCann and Geoghegan: New York, \$12.95.)

MOST of the generals of World War II published their memoirs in the 1950s. However interesting these are as accounts of events and as revelations of personality, they are pervaded by unreality because their writers could not reveal the extent of their previous knowledge of the enemy's plans and arrangements. Now enough has been declassified to enable a rational account to be given of the controversies that raged before major decisions were taken. The relaxation of the restrictions has led, recently, to the publication of several books of great interest, those by F. W. Winterbotham and by Solly Zuckerman being particularly informative. Now we have a book by R. V. Jones, the grand panjandrum of the arcana of scientific intelligence.

At Oxford in the 1930s Jones became a pupil and protégé of F. A. Lindemann (later Lord Cherwell). As a graduate student he worked on the development of instruments for the detection of infrared radiation and became involved in Lindemann's wrong-headed crusade to promote infrared detection as an alternative to radar. Jones escaped from this at the end of 1938 and started on what was to be his main work until 1946: the understanding of German equipment, activities and military doctrine.

He kept his connection with Lindemann throughout the War and made good, but restrained, use of the access it gave to Churchill. The myth that he could call down wrath from on high served him even better than the reality. I remember, for example, an occasion when an Assistant Director of Air Intelligence told me that he was going "to get rid of that man Jones"; I told him he couldn't and next day found a sadder and a wiser Air Commodore. Jones was not universally popular; perhaps this was inevitable as he was attacking strongly held beliefs and was usually proved to be right.

When it became clear that he was right he sometimes did not soften the blow to his opponent's pride. He was given to practical jokes and to contriving situations in which they looked foolish. The story of the lump of coke that was said to have fallen from a Russian pilot-less aircraft and to contain 98% of an element 'unknown to science' is an entertaining example.

Lindemann's support of Jones was probably the most useful thing he did during the War. Since he died his reputation has suffered from attacks of unexampled virulence. The latest is J. Z. Young's chiding of Solly Zuckerman for not recognising "what an odious man he was" (*Nature* 271, 387; 1978). Was he odious? Readers of Jones' book can judge for themselves; for me it strengthens the impression that Lindemann suffered from an almost pathological unsoundness of judgement; if two views were possible, he almost invariably supported the wrong one. He was not an easy or a likeable man, and one of his least amiable traits was a deep and unrelenting hatred of the German people, even to the extent of omitting from *Who's Who* the fact that he was born in Germany. His support of Jones was, however, effective and important; it is interesting to speculate whether Tizard or Blackett would or could have done it as well if they had been in Lindemann's place.

Jones' first great success was the penetration of the system of radio-beams that enabled the Germans to bomb cities in England by night. This not only enabled the system to be rendered ineffective by jamming and by "bending the beams" but also drew attention to the gross neglect of navigational aids by our own bomber force who, at the start of the war, believed that they could successfully attack targets deep in Germany and needed no new aids.

Jones' success with the beams, with the understanding of German radar and, later, with the German flying bombs and rockets depended on the intimate connections he maintained with his sources; with the photo-interpretation unit at Medmenham, with the briefing of agents on the continent and with the cryptographers at Blech-

ley Park. No-one else had the overall grasp of what the sources could and could not provide. To me his most impressive achievement was the discovery and elucidation of the radio-signals that gave the tracks of flying bombs and rockets fired on the test range along the Baltic. In this he can truly say with the Duke of Wellington: 'I don't think it would have done if I had not been there'.

The book is a wonderful story of what a young man (he was 28 in 1939) can do. Like many others he knew that the things he did then were the most important things he would ever do and, now that he at last comes to explain it all to a wider audience, he is sometimes, perhaps, unnecessarily insistent on the follies of his opponents: though some of them were, indeed, remarkably foolish.

The accounts of practical jokes and subterfuges for outwitting politicians and Air Marshalls should not obscure the important lessons of this book. A government that takes action on irrationally alarmist views concerning another country's intentions or capabilities may precipitate a disaster. Such judgments depend heavily on the views developed by the intelligence organisations, of which scientific intelligence is an important component. Jones' main conclusions are that, although collecting information needs large numbers of people, the group that develops the meaning of the information should be small, should have close relations with the sources and should have as wide a range of experience as possible. His warning against leaving such matters to those working on our devices is of special importance. They are too close to our own problems and may conclude that, because a device, such as the V-2, is a poor choice for us, the Germans cannot be making it; and the evidence that they are must be a hoax.

This book provides an insight into the processes of intelligence. Whether this is desirable is dubious. The Russians know it all, but do the Cubans, the Cambodians or the Ethiopians? □

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Laser and microwave techniques

Laser and Coherence Spectroscopy.
Edited by Jeffrey I. Steinfeld. Pp. 530.
(Plenum: New York, 1978.) \$54.

THE application of coherent transient techniques such as free-induction decay, optical nutation and photon echoes in molecular spectroscopy is providing new information and fresh insights into basic molecule-radiation interactions and collisional and radiative relaxation processes. This book contains five chapters by separate authors, mainly chemists, on laser and microwave techniques currently in use.

The opening chapter by Steinfeld and Houston concentrates on methods and devices used in double resonance and is followed by a chapter on coherent transient microwave spectroscopy by Schmalz and Flygare. The third chapter by Shoemaker on coherent transient infrared spectroscopy emphasises the Stark switching method he pioneered with Brewer. The fourth chapter by Harris and Breiland is concerned with coherent spectroscopy in electronically excited states, and the book concludes with a chapter by Novak Friedman and Hochstrasser entitled "Resonant Scattering of Light by Molecules: Time dependent and Coherent Effects".

Two chapters are worth singling out for special mention. Shoemaker's lengthy chapter contains a quite outstanding analysis of the optical Bloch equations using Jaynes' elegant matrix method of solution, and all his theoretical arguments are prefaced by useful pedagogical or heuristic explanations. I particularly liked his discussion of phase interruption and velocity-changing collisions.

The chapter by Novak, Friedman and Hochstrasser is noteworthy for other reasons: the authors seem unaware of some of the advances made in quantum optics in the past 20 years and step into traps I thought had rusted away years ago. They use concepts such as a "nearly monochromatic photon", and "photons of variable extents in time", and make mis-statements (p 457) on the preservation of coherence when a molecule interacts with a coherent state field, and on the nature of chaotic light (p 473), and so on.

Altogether, the book is an odd mixture. Specialists should read it, if only for Shoemaker's article. Textual evidence suggests the manuscript was completed in 1976. Given this delay and the extravagant price, the authors would have been better advised to publish their work in existing review journals or in an *Advances* series.

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Cellular recognition

Receptors and Recognition. (Series A.) Vols 1-4. Edited by P. Cuatrecasas and M. F. Greaves. (Chapman and Hall: London; Halsted: New York, 1976-1978.) Vol. 1: £7, £4.50; vol. 2: £10, £6; vol. 3: £10, £6; vol. 4: £11.50, £7.50 (hardback and paperback, respectively).

CELLULAR recognition processes are now the subject of a vast research effort, but most of this is compartmentalised into rather narrow specialities. As pointed out by the editors of this series (a biochemist and an immunologist), any attempt to reveal underlying unities in the various types of cellular recognition should consider processes as diverse as "fertilisation, embryonic development, infectious interactions, the activity of the nervous system, the regulation of growth and development by hormones and the immune response... to antigens". Given this large canvas on which to work, it is inevitable that any editors' choice of topics will be somewhat personal, but one hopes for a balance to be struck between descriptions of biological phenomenonology and of molecular mechanisms and for each constituent subject to get fair representation.

Based mainly on these four volumes, but also on the titles of contributions to two further volumes which will appear this year (due June and September), I feel that the editors of this multidisciplinary series are to be congratulated on progress so far. However, they do not seem to have yet achieved quite the necessary balance between different areas, perhaps because they have allowed too much 'background' material to occupy valuable space which could have been better used.

This imbalance in the first four volumes tends to favour immunological topics and discriminate against topics related to mammalian homeostasis and tissue function. The immune system is the central topic in *The Evolution of Receptors and Recognition in the Immune System* (F. M. Burnet, Vol. 1), *A Structural Basis for Molecular Recognition: The Antibody Case* (D. Givol, Vol. 2), *Cell Traffic* (M. A. B. de Sousa, Vol. 2) and *The Cellular Receptor for IgE* (H. Metzger, Vol. 4). In addition, it features prominently in *Membrane Associated Events in Lymphocyte Activation* (K. Resch, Vol. 1), *Specificity in Host-Parasite Interactions* (K. N. Brown, Vol. 1), *Calcium and Cell Activation* (B. D. Gomperts, Vol. 2), *Antibodies to Receptors for*

Acetylcholine and other Hormones (J. Lindstrom, Vol. 3) and Endocytosis (T. P. Stossel, Vol. 4). There is also an emphasis on cellular compatibility and incompatibility in the treatments of Incompatibility in Flowering Plants (D. Lewis, Vol. 2), Mating-type Interactions in Microorganisms (M. Crandall, Vol. 2) and Virus Receptors (A. Meager and R. C. Hughes, Vol. 4).

By contrast, there are only three articles the primary emphasis of which is on receptors for hormones or neurotransmitters: Catecholamine Receptors (A. Levitski, Vol. 2), Acetylcholine Receptors (M. E. and A. T. Eldefrawi, Vol. 4) and Hormone Action at the Plasma Membrane: Biophysical Approaches (M. Sonenberg and A. S. Schneider, Vol. 4), with additional material in the Lindstrom article.

So far, the only general treatment of polypeptide hormones is in the Sonenberg and Schneider contribution. This is a curious chapter which attempts, I think unsuccessfully, to unify all aspects of hormone action into a single 'integrated model'. Some correction of the bias against hormones may come with forthcoming chapters on Relationships in the Structure and Function of Receptors for Glycoprotein Hormones, Bacterial Toxins and Interferon (L. Kohn, Vol. 5) and Cyclic Nucleotides (J. Fain, Vol. 6).

Chapters which may be regarded as background include a description of antibody structure and specificity which forms much of Givol's article (Vol. 2), Erythrocyte Proteins (H. Furthmayr, Vol. 3) and Specificity of Membrane Transport (M. Silverman, Vol. 3). In the future, we are promised Stereoselective Molecular Recognition in Biology (R. A. Lehman, Vol. 5), Fluorescence and NMR Studies of Membranes (A. G. Lee, Vol. 5) and Reconstitution of Biological Membranes (G. Eytan and B. Kanner, Vol. 6).

The remaining articles are a good introductory discussion entitled Cell Surface Receptors: A Biological Perspective (M. F. Greaves, Vol. 1) and two topics announced for Vol. 6: Rhodopsin: A Light-sensitive Glycoprotein (P. O'Brien) and Hepatic Degradation of Circulatory Glycoproteins (G. Ashwell). Referring back to the areas which the editors recognised as within the scope of the series, it becomes obvious that nothing has yet been seen of cell-cell interactions in morphogenesis and in tissue differentiation.

In general, the contributions to the series are of high quality. Most of the authors have managed to make them comprehensible to a general audience, but in a few cases the uninitiated may be bamboozled by barrages of unexplained technical terms (for example, ectoperitrophic, metacyclic, proventricular, schizont and anamnestic all come within a few pages in Brown's article). It is a pity that the editors did not tidy up the inelegant writing which occasionally mars most contributions, and that few chapters seem to have been carefully proof-read. This untidiness is accentuated by a unjustified style of typesetting. These volumes have no indexes, but a cumulative index is promised for Volume 6. Although this may be appropriate for libraries (until someone steals Volume 6), it will give little comfort to the individual buyer who only wants part of the series.

In summary, therefore, this is a good series which can help to provide the 'broader education' needed by workers in the multidisciplinary world of cellular recognition. With some further work on the editorial and production side, future volumes might cause this verdict to be modified to excellent. **Bob Michell**

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Chapter 8 describes in detail the photochemistry of the polluted troposphere and introduces in a useful way for the uninformed reader the elements of photochemical theory. Chapters 9-11 deal with the chemistry of the stratosphere and mesosphere, and especially highlight the possibility of changes in the ozone concentration arising from the effects of supersonic transports and the release of chlorofluorocarbons. Considering the very rapid developments in this field these chapters are well up to date.

The early chapters of the book must be read with caution; they are unfortunately marred by a number of misleading statements. For instance, on p 16 the dominant role of collisions in establishing local equilibrium between the radiation and thermal field in most of the atmosphere is ignored; on p 44 the CO₂ 15 μm band is stated to be invariably in local thermodynamic equilibrium; and on p 21 the important absorption by water vapour in the Earth's atmosphere is ignored.

The chapters on atmospheric chemistry are the most useful parts of the book; they are particularly appropriate for students and research workers in the field of atmospheric pollution. Furthermore, the book is moderately priced and well produced. **J.T. Houghton**

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Atmospheric physics and chemistry

Energy and the Atmosphere: A Physical-Chemical Approach. By Ian M. Campbell. Pp. 398. (Wiley: Chichester, Sussex, New York and Sydney, 1977.) Hardback £14.50, \$31; paperback £5.95, \$12.50.

THIS book is concerned with two topics, the chemistry of energy generation and chemical processes in the atmosphere. The first three chapters serve as an introduction to the gross structure of the atmosphere, especially to the way in which solar radiation is modified on its passage through the atmosphere to the Earth's surface. The next three chapters deal with the photosynthetic origin of fuels, combustion, and briefly the thermodynamics of heat engines and fuel cells.

The last four chapters, which make up over half the book, deal with various topics in atmospheric chemistry.

In chapter 7 a summary is given of the major chemical cycles of the atmosphere, namely the hydrogen, carbon, oxygen, nitrogen and sulphur cycles. Only a brief mention is made of the problem of increasing atmospheric CO₂ and its possible effect on climate through the enhanced greenhouse effect. The reader is left with little feel for the present state of knowledge of the likely rate of CO₂ increase as the consumption of fossil fuels increases; problems such as the exchange of CO₂ with the oceans is only dealt with very cursorily. Considering the title of the book and the fact that the CO₂ problem is likely to feature in an important way in the energy debate of the next decade or two, a much more detailed account of this problem would have been appropriate.

Physics of nuclei and elementary particles

Nuclei and Particles: An Introduction to Nuclear and Subnuclear Physics. By Emilio Segré. Pp. 966. (W. A. Benjamin: Reading, Massachusetts, 1977.) \$29.50.

THE original intention of this well known book, first published in 1964 and now in a second edition, remains unchanged, namely to provide potential research workers with a broad, but not superficial, introduction to the physics of nuclei and elementary particles, paying attention both to the theoretical aspects of the underlying physics and also to the experimental methods and techniques used in this field. The theoretical treatment is kept at the lowest possible level of sophistication necessary for a good understanding of the physical processes under discussion, and much healthy use is made of intuitive argument. Even so, a sound knowledge of quantum mechanics is expected of the reader and occasionally a familiarity with the elements of relativistic quantum mechanics.

Apart from a small introductory section and a set of appendices dealing with various technical matters, the book is divided into three main parts: "Tools", "The Nucleus" and "Particles", occupying respectively 184, 469 and 239 of the total of 966 pages.

The first part is essentially in the same form as in the first edition, although thoroughly updated and with the inclusion of one or two additional subsections dealing with recent developments. There is a particularly fine and meaty chapter on the passage of radiations through matter, together with chapters on detection methods, particle accelerators (including a new section on colliding beams) and on the general features and applications of radioactive decay. Plenty (but not in excessive quantity) of technical detail is given and this part of the book, in particular, is written with masterly authority.

The discussion of what is generally called low energy nuclear physics in the second part of the book, covers the ground one would expect. There are large chapters on nuclear structure and nuclear reactions and smaller ones on α , β and γ decay, the two-nucleon interaction, and neutron physics. Here, the balance and ordering of the matter dealt with might be questioned. For example, in a book of this size which sets out to be "introductory", to devote only seven pages (including three full pages of diagrams) to the nuclear shell model which, after all, underlies a lot

of our thinking about the nucleus, seems rather parsimonious. It could also be argued that a discussion of the nuclear force itself and the two-nucleon system would better precede the treatment of nuclear forces in the nucleus, in particular, the role of the residual interaction and pairing effects. But these are minor points which hardly detract from what is an excellent treatment of the core of nuclear physics.

It is in the part of the book dealing with elementary particle physics that major differences between the two editions are to be found. This section has been completely rewritten and Professor Segré has now provided a wide, comprehensive and up-to-date account of the state of play in this field. After a general introductory chapter dealing with some general symmetry and conservation laws, he devotes succeeding chapters to leptons, pions, and other bosons and baryons. There follows a major chapter on the SU(3) classification of hadrons and quarks, with discussion of charm and colour. No attempt is made to deal with the mathematical details of the SU(3) group which are regarded (rightly) by the author as "beyond the scope of this book", a phrase which appears more than once in these last two hundred or so pages. The remaining three chapters deal with e^+e^- collisions (including discussions of the J/ψ particle), weak interaction physics (including unified theories) and CP violation, and high energy collisions of hadrons. Inevitably, because of the extreme mathematical complexity of much of the underlying theory, the treatment in this last part of the book has to be carried out at a more superficial level. Despite this limitation, the author has

managed most successfully to deal in an understandable way with a great many of the current ideas of elementary particle physics in line with his intention to introduce the reader to as many trees in the forest as possible.

Layout and typography are much to be preferred to those of the first edition, the layout of which was odd in the extreme. Particularly striking are the many diagrams, both illustrating apparatus and representing quantitative data in graphical form. Most are accompanied by excellent and full captions, and are extremely informative. As befits a pedagogical book, each chapter is accompanied by a set of problems which vary from simple numerical calculations to effective extensions of the subject matter of the chapter. In addition there is a general bibliography, a bibliography for each chapter and plentiful referencing to original papers.

The book is quite unique and constitutes a remarkable and readable exposition of most of the basic stuff of nuclear and particle physics ranging from details of experimental method through to advanced theoretical ideas. The whole is illuminated and characterised by the author's physical insight and admirably achieves his aim of conveying an intuitive understanding of the main phenomena of nuclear and particle physics to "physics students, chemists, and engineers who want to acquire enough knowledge of nuclear and subnuclear physics to be able to work in this field". Finally, it is not expensive for its size.

R. J. Blin-Stoyle

R. J. Blin-Stoyle is Professor of Theoretical Physics at the University of Sussex, UK.

Nature of the seabed in a median valley

Photographic Atlas of the Mid-Atlantic Ridge Rift Valley. By R. D. Ballard and J. G. Moore. Pp. 114. (Springer: New York and Berlin, 1978.) DM43.30; \$21.70.

It is always difficult to publish for the benefit of one's fellow scientists observations that are entirely visual and very numerous. Ballard and Moore have produced an excellent book summarising the results of surveys by underwater photography, both from the submersible ALVIN and from the US Navy LIBEC System, which were made in support of the international study of accretion processes on the axis of the Mid-Atlantic Ridge, south-west of the

Azores (Project FAMOUS). The features shown in the many thousand photographs taken, have been categorised into two classes: first, submarine volcanic products including the vents themselves, flow units and a great variety of pillow lavas and breccias; and second, faults, fissures and related tectonic features. As well as describing the geological setting, interpretations of the photographs are illustrated by excellent sketches and by diagrams showing the processes creating the micro-topography.

I thoroughly recommend the book not only as a serious scientific record of the nature of the seabed in a median valley, but as an attractive and interesting book for the non-specialist.

A. S. Laughton

A. S. Laughton is Director of the Institute of Oceanographic Sciences (NERC), Wormley, UK.

obituary

C. A. J. Young

MR C. A. J. YOUNG F.R.S., died on 20 January 1978 at the age of 65. Mr Young (Christopher at home but A. J. at work) was the fourth President of the Institute of Measurement and Control from 1954 to 1957 and a recognised authority in that area of technology. He was head of the ICI Central Instrument Research Laboratory for some 25 years and the author of a definitive textbook on the fundamentals of automatic process control.

His achievements were recognised by the award of an Honorary D.Tech. by the University of Bradford in 1968, the award of the first Sir Harold Hartley medal by the Institute of Measurement and Control in 1969 and Honorary Fellowship of that institute in 1973. In 1972 he became a Fellow of the Royal Society.

Born on 7 March 1912, he read physics at Oxford and after taking his BA in 1932 he worked for a year in Professor Townsend's research group, gaining his B.Sc. for work on electrical discharges in gases at low pressures. He then turned to teaching and was a master at Cheltenham for some four years before joining the Meteorological Service in 1938. This work took him to the Sudan where he was involved in the establishment of meteorological stations, particularly to support the African services of Imperial Airways. He returned to England at the outbreak of war, hoping to join Coastal Command as he already had some flying experience; but the Air Ministry perceived a more effective use for a scientifically qualified meteorologist as part of their joint research project with ICI for fog dispersion on airfields (FIDO).

In 1940 he joined the Chemical Engineering Research Section of ICI (Fertiliser and Synthetic Products) Ltd at Billingham, his first major involvement with instrumentation. He joined the Billingham 'Tube Alloys' nuclear engineering team on its inception and found himself responsible for the instrument side of the project. From then on instrumentation and control became his main interest, particularly in relation to chemical engineering activities, and he was to work with ICI in this field until his retirement in 1973.

He left Billingham in 1946 to work in the Technical Department of the ICI Head Office in London. The Technical Director, F. E. (later Sir

Ewart) Smith, made him responsible for establishing the ICI Central Instrument Section with a small laboratory facility at The Frythe in Welwyn, Herts.

This began the most productive part of his career. A. J.'s greatest strengths lay in his ability to recognise technical areas where exploration could be fruitful, to select good lieutenants, to listen to them, to give them freedom and support, and to enlist the support of unbelievers. The new laboratory was quickly established with great enthusiasm and, it seemed, with a complete disregard of the procedural obstacles that usually characterise large organisations. There was early work on the development of techniques for on-line analysis and quality control but the work of the laboratory soon focused on the substitution of understanding for empiricism in the application of automatic process control. The next few years showed a steady growth and by 1956 the group had outgrown its Welwyn facilities and moved to Bozedown House near Pangbourne in Berkshire. By this time its name was well-established. The laboratory and A. J. with it had developed an international reputation and saw a steady stream of visitors from Britain and overseas who regarded it as a major centre of process control technology.

Within a short time after the move to Bozedown House A. J. Young established the first digital computing service at Wilton which led to the introduction of computers all through the Company and again, it was his first computer-controlled pilot plant which led to the installation at the Alkali Division at Fleetwood of one of the first large scale computer-controlled plants in the world.

Following an ICI Head Office reorganisation, what had been an independent Central Instrument Research Laboratory was amalgamated with the Petrochemical and Polymer Laboratory to form the new 'Corporate Laboratory' and A.J. became Technical Director of the Corporate Laboratory—Bozedown. After his retirement in 1973 the new laboratory was to be unified with a new and broader emphasis, in a single location at Runcorn in Cheshire, thus ending an era which his friends and colleagues will always associate with their memories of him.

A. J.'s final illness was relatively short but over recent years he must have suffered much pain and his

friends will not forget his courage in disregarding it to the end.

We extend our sympathy to his widow and stepdaughters.

J. R. Halsall
J. D. Tallantire

J. W. McLeod

EMERITUS Professor James Walter McLeod, O.B.E. F.R.S., F.R.S.E., F.R.C.Path., born on 2 January 1887 at Dumbarton, died in his 92nd year on 11 March 1978.

After graduating from Glasgow Medical School in 1908 he held a Coats, and later a Carnegie scholarship in Robert Muir's Department of Pathology. From 1912 he was Assistant Lecturer in Pathology at Charing Cross Hospital Medical School until he joined the R.A.M.C. at the outbreak of war in 1914. At the end of war he was Captain in charge of the 8th Mobile Laboratory in France and had published papers on trench fever and nephritis, bacillary dysentery and the incidence of Pfeiffer's influenza bacillus in pandemic influenza. In 1919 he became Lecturer in Bacteriology in Leeds and in 1921 the first occupant of the Brotherton Chair in Bacteriology, which he held until his retirement in 1952.

From his war experiences stemmed his interest in anaerobic gas gangrene bacilli, and from this in turn his pre-occupation with bacterial respiration. In a series of 14 papers published between 1921 and 1929, mostly in collaboration with J. Gordon and sometimes others, with only a few exceptions published in the *Journal of Pathology and Bacteriology*, he explored the reducing mechanism of aerobic and anaerobic microorganisms, the production of H_2O_2 by anaerobic bacteria and by aerobic bacteria devoid of catalase and the colour changes they produced in haemoglobin or in benzidine blood media under these conditions. Two of these papers dealt with the oxidase reaction of bacterial colonies. McLeod's simplified technique and the second stage development of a bluish-black pigment by gonococcus colonies observed when *p*-phenylene diamine was used as indicator dye in the oxidase reaction so revolutionized the laboratory diagnosis of clinical gonorrhoea that he achieved an international reputation. The work on bacterial respiration culminated in McLeod's contributing chapters on the

subject to Jordan and Falk's *Newer Knowledge of Bacteriology and Immunology* in 1928, and to the Medical Research Council's *A System of Bacteriology* in 1930.

Having used an improved heated blood agar plate, better known as chocolate agar, for culturing gonococci McLeod then included a critical concentration of potassium tellurite, previously described by Conradi and Troch in 1912, in this medium for the selective growth of *Corynebacterium diphtheriae*. This combination of a much improved growth medium and an efficient selective agent made possible the recognition of two, and later three distinct colonial types of diphtheria bacilli. McLeod and his team soon realized that these three distinct colonial types were related to differential growth characters in nutrient broth and to type-specific polysaccharide and sugar fermentation reactions. From these observations McLeod's concept of these three biotypes being linked to three different degrees of clinical severity of the infection was born and he named these biotypes accordingly, i.e. *gravis*, *mitis* and *intermedius*.

The first paper by the Leeds team in 1931 triggered a flood of research effort by many bacteriologists in all parts of the world and kept the Leeds Department busy until the incidence of clinical diphtheria had declined so much that work came to a halt for lack of material and interest. Finally in 1950 McLeod validated his concept of the relation of the three main *C. diphtheriae* biotypes to clinical severity in an authoritative survey of reports on 25,000 cases from all corners of the globe, which was published in the *Journal of Pathology and Bacteriology*.

The main currents of McLeod's research activities were interspersed with papers on post-operative tetanus infection arising from soil dust in a surgical theatre, the prophylactic value of some antiseptics in experimental streptococcal wound infections, the carriage of *Bord. pertussis* in contacts of clinical cases, the differential laboratory diagnosis of *Cl. oedematiens* and the factor in nutrient agar which inhibits the antibacterial action of sulphonamides.

After retirement McLeod, somewhat reluctantly, had a two-year period of research inactivity but then began investigations in an Edinburgh hospital and for the Scottish hospitals Endowments Research Trust into the causes, reservoirs of, and urinary tract infections caused by *Ps. aeruginosa* in paraplegics and was able to propose some measures of prevention. When the grant ceased he was given research hospitality in the Edinburgh Central Microbiological

Laboratories where he participated in a series of investigations, published in the *British Medical Journal* in 1967, that provided the foundation for quantitative cellular and bacterial urine examination in newborns, infants and young children.

J. W. McLeod is survived by his second wife Joyce, a son and five daughters with whom all former associates share a great sense of loss.

K. Zinnemann

J. A. Douglas

PROFESSOR J. A. Douglas, D.Sc., Emeritus Professor of Geology and Palaeontology in the University of Oxford, died on 27 February 1978, at the age of 93.

James Archibald Douglas was born at Ilkley, Yorkshire on 1 December 1884. He was educated at Haileybury and Keble College, Oxford which he entered in 1902 as Open Scholar in Zoology. He took a First Class Honours in Geology in 1905 and, with the award of the Burdett-Coutts University Scholarship, began research under the versatile professor, W. J. Sollas. Douglas's first work was on the changes of physical constants which take place in certain minerals and igneous rocks, on the passage from the crystalline to the glassy state and he soon became involved in Sollas's ever-changing research interests which ranged from serial sectioning of fossil vertebrates to palaeoliths. He also began taking over at an early age an increasing amount of the undergraduate teaching. Nevertheless, Douglas spent several vacations in Ireland investigating the Carboniferous Limestone rocks of Co. Clare.

In 1910, he set out on a two years' geological exploration of the Andes in Peru and Bolivia at the suggestion and expense of W. E. Balston, a graduate of Oxford and generous benefactor of the University Museum. He finally completed this work after a return visit in 1929.

In August 1914, Douglas was commissioned and was on active service in France throughout the War, being wounded in 1915. On his return to France, he was seconded for mining duties and was responsible for the large mines at St Eloi, Messines Ridge in 1916, where he was wounded for the second time. In 1917 his corps of miners constructed the Grange Tunnel for the attack on Vimy Ridge.

When Douglas returned to Oxford in 1919, his chief was already seventy years of age, and although Sollas was to remain in control of the department for another seventeen years, he became increasingly remote and un-

predictable, with interests that were almost exclusively anthropometric and archaeological. On Douglas almost exclusively, fell the task of coping with an overwhelming influx of ex-service students, not only honours men on shortened courses but engineering students and those destined for the Colonial forestry service. Everything had to be done on a very limited budget and constant improvisation, but Douglas's reward was in the friendships he then established with men destined to scatter to responsible posts throughout the world.

At this time, he began an association with the major petroleum companies and he was for thirty years Palaeontological Adviser on the Advisory Board of the Anglo-Iranian Oil Company, (British Petroleum Company). During this time most of the palaeontological material collected by the company's exploration staff was examined by him and the resulting reports and correlations incorporated his identifications.

In 1937, he succeeded to the Oxford chair and at once was able to carry out his long made plans for a revitalising of Oxford geology teaching and research. He obtained a substantial donation from Shell towards a new departmental building, and the plans were well-advanced in 1939. During the second War he was Town Defence Commander for Oxford.

The new Geology department was the first science building to be erected in Oxford after the war but already the post-war enlarged intake of students had been repeated and with it the need for improvisation and economy. Douglas had the satisfaction, however, of doing his last year of teaching in the new building. On retirement in 1949, his break with the department he had served so long was almost complete, for he moved to Yarmouth, Isle of Wight. There he indulged his favourite pastime of sailing, racing his own yacht in competitions for many years, enjoying as he did perfect health till near the end of his long life.

Douglas was a joint secretary of the Geological Society of London with W. Campbell Smith from 1922-1929, and served on its Council for a total period of almost twenty years. His industry and achievements were not however of a type which readily attracts public awards and indeed he received few of these. He set far more importance on the satisfaction he received from the real affection and gratitude expressed by generations of former students most of whom moved into academic and industrial posts of responsibility across the world, and remained in touch with him as life-long friends.

James M. Edmonds

announcements

Awards

The Chemical Society 1978 Pedler Lectureship has been awarded to **Professor Sir James Baddiley**, Director of the Microbiological Chemistry Research Group of the University of Newcastle upon Tyne; and the 1978 Tilden Lectureship to **Professor J. J. Turner**, Head of the Inorganic Chemistry Dept of the same university.

The London Mathematical Society has awarded the third biennial Whitehead Prize to **Professor Ioan Mackenzie James, FRS**, Savilian Professor of Geometry in the University of Oxford, for his work in algebraic topology, particularly in homotopy theory.

The Institution of Mining and Metallurgy has made the following awards: **Mr R. H. MacWilliam, OBE**, has been awarded the Gold Medal of the Institution for 1977 for outstanding services to the Institution and to the minerals industry internationally; **A. L. Thomas, OBE**, has been made an Honorary Fellow for distinguished services to the Camborne School of Mines and to the mining industry in Cornwall; **J. W. Norman, N. J. Price**, and **E. R. Peters**, have jointly been awarded the Consolidated Gold Fields Limited Gold Medal and Premium of £300 for their paper entitled 'Photogeological fracture trace study of controls of kimberlite intrusion in Lesotho basalts'; **A. G. Moncrieff**, and **P. J. Lewis**, have jointly been awarded the Consolidated Gold Fields Limited Silver Medal and Premium of £200 for their paper entitled 'Treatment of tin ores'.

The Royal Society of Victoria Research Medal for 1977 has been awarded to: **Professor R. D. Brown**, Professor of Chemistry, Monash University, for studies on molecular structure by the application of quantum mechanics and of microwave spectroscopy, and in radio astronomy leading to the discovery of new interstellar molecules. **Dr W. F. Budd**, Senior Glaciologist, Antarctic Division, Department of Science, for his work on the dynamics of ice masses and the flow and surging of glaciers. **Professor B. H. J. McKellar**, Chairman, School of Physics, University of Melbourne, for his work in theoretical nuclear physics involving both nuclear and particle theory.

The International Meteorological Organization (IMO) Prize, which is awarded annually for outstanding work in meteorology and international collaboration, was awarded by the WMO Executive Committee to **Dr Alf E. G. E. Nyberg**, former Director General of the Swedish Meteorological and Hydrological Institute.

The Appleton Prize for Ionospheric Physics for 1978 has been awarded by the Royal Society to **Professor P. M. Banks**, head of the department of physics at Utah State University, Logan, for his studies of the plasma flow between the ionosphere and the magnetosphere.

The 1978 Hewlett-Packard Europhysics Prize has been awarded to **Professor Zhores Ivanovich Alferov** of the A.F. Ioffe Physico-Technical Institute in Leningrad for his outstanding contribution to the development of a new type of semiconducting material which can be used to produce, for example, highly efficient solar cells and lasers with a continuous output.

The Geological Society of London has announced the following awards for 1978: The Wollaston Medal to **Professor J. Tuzo Wilson** for major contributions to the evolution of the concept of sea floor spreading and continental drift; The Murchison Medal to **Dr Stephen Moorbath** for contributions to knowledge of crustal history, through the development and application of isotope studies; The Lyell Medal to **Dr R. G. C. Bathurst** for his contribution to knowledge of carbonate sediments and their diagenesis; The William Smith Medal to **Dr M. King Hubbert** for his work on petroleum geology and on hydrodynamic conditions and fluid behaviour in petroleum reservoir engineering; The Wollaston Fund, Murchison Fund and equal moieties of the Lyell Fund to **Dr D. J. J. Kinsman**, **Dr W. D. Ian Rolfe**, **Dr Paul L. Hancock** and **Dr A. W. A. Rushton**.

The George Fleming Prize for 1977 has been awarded to **Mr R. J. Esslemont** of the Department of Agriculture, University of Reading, and **Mr R. G. Eddy** in practice at Shepton Mallet, Somerset, for their article *The Control of Cattle Fertility: The Use of Computerised Records*. This annual prize is for the best article from the *British Veterinary Journal* in the preceding year.

Professor Isaak Wahl of the Tel Aviv University has been awarded the Harvey Prize in Science and Technology for his research into the improvement of cereal grains.

The International Organisation of Plant Breeders for the Protection of Plant Varieties, ASSINSEL, have created a new award for scientists whose basic research work has made a considerable contribution to the improvement of plant breeding methods for the benefit of agriculture and horticulture. This Award will be granted every four years beginning with 1978, **Mr André C. Gallais** from France being the first recipient for his thesis based on observations with lucerne (alfalfa), a biometrical study on heterosis in an allogamic autotetraploid species.

Appointments

Dr J. Paul Wild, 55, is to be the new chairman of the Commonwealth Scientific and Industrial Research Organization (CSIRO) for seven years from 25 September 1978.

Professor J. J. A. Reid, Chief Medical Officer of the Scottish Home and Health Dept, Edinburgh has been elected chairman of the World Health Organization's Executive Board.

Royal Irish Academy: president and officers for 1978: president: **Professor G. F. Mitchell**, Professor of Quaternary Studies, Dublin University. Treasurer: **Professor P. Lynch**, Professor of Political Economy, University College, Dublin. Secretary: **Professor J. C. I. Dooge**, Professor of Civil Engineering, University College, Dublin. Science Secretary: **Professor M. C. Sexton**, Professor of Electrical Engineering, University College, Cork. Polite Literature and Antiquities Secretary: **Dr M. de Paor**.

The Institution of Chemical Engineers president for 1978-79 is **John Solbett**, a Director of Humphreys and Glasgow Ltd, who succeeds J. R. S. Morris, Deputy Chairman of the National Enterprise Board. The two new vice-presidents are **Dr A. W. Pearce**, Chairman of Esso Petroleum Ltd, London; and **Professor W. L. Wilkinson**, Professor of Chemical Engineering in the University of Bradford.

The National Academy of Sciences today announced the election of 15 scientists from nine countries as foreign associates of the Academy: **Michael F. Atiyah**, Royal Society research professor, Mathematical Institute, Oxford University, UK; **Ricardo Bressani**, chief, division of agricultural and food science, Institute of Nutrition of Central America and Panama, Guatemala City, Guatemala; **Luigi L. Cavalli-Sforza** (Italy), professor of genetics, Stanford University, Stanford, California; **John W. Cornforth**, Royal Society research professor, department of chemistry, University of Sussex, UK; **James H. S. Gear**, consultant, Institute for Virology, South African Institute for Medical Research, Sandringham, Republic of South Africa; **Johannes Geiss**, professor of physics and director, Institute of Physics, University of Berne, Switzerland; **Robert A. Hinde**, honorary director, Medical Science Research Council Unit on Development and Integration of Behaviour, and Royal Society research professor, University of Cambridge, UK; **Hugh E. Huxley**, deputy director, Medical Research Council Laboratory of Molecular Biology, Cambridge, UK; **Helge Larsen**, Royal Society research professor, department of ethnology, Danish National Museum, Copenhagen, Denmark; **Rudolph L. Mossbauer**, professor of physics, Technical University, Munich, Federal Republic of Germany; **Joseph Needham**, master (retired), Caius College, Cambridge, UK; **Giuseppe Occhialini**, professor of physics, Institute of Physical Science, University of Milan, Italy; **John C. Polanyi**, university professor and professor of chemistry, University of Toronto, Canada; **Maarten Schmidt** (Netherlands), chairman, division of physics, mathematics, and astronomy, California Institute of Technology, Pasadena, California; **Rudolph Trümpy**, professor, Geological Institute, Eidgenössische Technische Hochschule, Zurich, Switzerland.

The Natural Environment Research Council has appointed **Dr Anthony S. Laughton** as Director of the Institute of Oceanographic Sciences (IOS) to succeed **Professor Henry Charnock, FRS**, who has taken up the Chair of Physical Oceanography at the University of Southampton.

The Director General of the International Atomic Energy Agency (IAEA) has announced that **Professor Johannes J. Gruemm** (Austria) has been appointed the new Deputy Director General for the Department of Safeguards.

Person to person

Biochemist visiting New Zealand offers 3-bedroomed furnished house at Wylam, near Newcastle upon Tyne, for one year from August 1978. Details from Dr C. N. A. Trotman, Department of Physiology, University of Newcastle upon Tyne.

Exchange 3-bedroomed fully-furnished centrally-heated home near London for similar home in Los Angeles convenient for U.S.C. and U.C.L.A., for 9 months or one year from late August 1978. London house 35-40 min from University College, British Museum, L.S.E. etc. Contact: E. A. Power, Department of Mathematics, University College, Gower Street, London WC1, UK.

Exchange 3/4-bedroomed furnished house in Dundee (2 miles Ninewells Teaching Hospital, 15 miles St Andrews) for similar in any pleasant location Europe conducive to writing, for some/all period July 1978 to January 6, 1979. Contact Nigel Harris, Philosophy Department, The University, Dundee, Scotland, UK.

Chemist seeks 3-4 bedroomed house, South West Minneapolis, U.S.A. September 1978-August 1979. Will exchange/rent own 4-bedroom house Kibworth near Leicester, England for same period. Contact Dr P. J. Craig, School of Chemistry, Leicester Polytechnic, PO Box 143, Leicester, UK.

We are preparing for publication letters written by J. Robert Oppenheimer before 1946. The principal archival collections have been consulted, and the edition is nearing completion. We would appreciate hearing from individuals who have, or know of, such letters. Contact: Alice Kimball or Charles Weiner, Massachusetts Institute of Technology 20D-224, Cambridge, Massachusetts 02139.

To all non-medical scientists in medical research: would any scientists (technical through to Professor grade) in the above category in hospitals or medical schools in the UK and all others deeply concerned about the problems relating to the absence of any permanent funding and career structure for the above please contact: Dr H. Anne Simmonds, Purine Laboratory, Clinical Science Laboratories, 18th Floor Guy's Tower, Guy's Hospital, London, UK.

Meetings

1-4 August, **27th Denver Conference on Application of X-ray analysis**, Denver (Mrs Mildred Cain, Denver Research Institute, University of Denver, Colorado 80208).

7-9 August, **9th International Conference on Photochemistry**, Cambridge (Cambridge Photochemistry Conferences, Dept of Physical Chemistry, Lensfield Road, Cambridge, UK).

8-9 August, **Mathematics and the Real World**, Roskilde (International Workshop Organising Committee, Roskilde University Centre, Post box 260, DK-4000 Roskilde, Denmark).

10-12 August, **2nd International Conference on the Photochemical Conversion and Storage of Solar Energy**, Cambridge (Cambridge Photochemistry Conferences, Dept of Physical Chemistry, Lensfield Road, Cambridge, UK).

13-17 August, **Fall Meeting of the American Society for Pharmacology and Experimental Therapeutics and Society of Toxicology**, Houston (ASPET-SOT, College of Pharmacy 141 SR-2, University of Houston, Texas 77004).

14-17 August, **Topics in the Biochemistry of Natural Products**, Oklahoma (Dr George Waller, Department of Biochemistry, Oklahoma State University, Stillwater, Oklahoma 74074).

16-23 August, **3rd International Congress of Plant Pathology**, Munich (Congress Plant Pathology, Biologische Bundesanstalt, Messeweg 11/12, D-3300 Braunschweig, FRG).

20-26 August, **4th International Conference on Animal Production**, Buenos Aires (General Secretariat, Reconquist a 533- 6°PISO, 1003 Buenos Aires, Argentina).

21-25 August, **Fundamentals of Noise and Vibration Control**, Massachusetts (Director of Summer Session, Room E19-356, Massachusetts Institute of Technology, Cambridge, Massachusetts 02139).

21-25 August, **Meteorology over the Tropical Oceans**, London (Royal Meteorological Society, James Glaisher House, Grenville Place, Bracknell, Berkshire, UK).

22-27 August, **Meeting on Transfer RNA**, Cold Spring Harbor (Gladys Kist, Cold Spring Harbor Laboratory, PO Box 100, Cold Spring Harbor, New York 11724).

22-31 August, **Infrared and Raman Spectroscopy of Biological Molecules**, Athens (Professor T. Theophanides, National Hellenic Research Foundation, 48 Bassileos Constantinou Avenue, Athens 501/1, Greece).

27-31 August, **International Conference on Behaviour of Offshore Structures**, London (BOSS 79, BHRA Fluid Engineering, Cranfield, Bedford, UK).